MEDICAL RESEARCH COUNCIL
Studies on the Nutritive Value of Bread and on the Effect of Variations in the Extraction Rate of Flour on growth of Undernourished Children.

by

Widdowson & McCance.

3247 (1) breads
(2) clinical examinations
(3) milk supplement
(4) animal experiments
(5) child height (10) skeletal
(6) " weight development
(7) mineral metabolism
(8) B vitamin " (11) child
(9) experimental feeding nutrition

Thms

PRIVY COUNCIL

MEDICAL RESEARCH COUNCIL SPECIAL REPORT SERIES
No. 287

Studies on the Nutritive Value of Bread and on the Effect of Variations in the Extraction Rate of Flour on the Growth of Undernourished Children

E. M. WIDDOWSON and R. A. McCANCE

LONDON: HER MAJESTY'S STATIONERY OFFICE
1954

MEDICAL RESEARCH COUNCIL

PREFACE

EARLY in 1946 Professor McCance and Dr. Widdowson visited the British Zone of Germany to report on the state of nutrition in that country, and later in the same year the Council set up a Unit at Wuppertal to investigate the effects and treatment of undernutrition. Two reports of the work of this Unit, which was under the direction of Professor McCance, have already been published in the Council's Special Report Series, Nos. 275 and 279, the first being mainly a study of the effects of undernutrition on the physiology of the individual, and the second an assessment of the value of foods of plant origin as the chief source of protein in the diets of young children. With the publication of the present volume the account of the Unit's main activities is completed.

The investigations described in this Report were carried out in two German orphanages on children whose previous diet had been insufficient for their needs and who were, in consequence, below the weight and height usually considered normal for their age. For the purposes of the trial, the children were divided into groups and given, in addition to their German rations, as much as they wanted of one of several specially prepared breads made from flours of different extraction rates. It was hoped that the children's growth and health during the subsequent year would reveal any major differences in the nutritional value of the various breads.

Perhaps the most striking observation was that these children grew even better than normal children of the same age in spite of the very simple diet provided, which contained little milk or other animal protein and had up to 75 per cent of its calorie value in the form of flour products. During the year for which the trial continued, growth was equally good on each of the experimental flours. It remains an open question, however, whether the flours were equally nutritious, or whether differences existed between them which were obscured by the exceptionally rapid growth-rate of undernourished children when receiving a diet adequate in calories.

Dr. Widdowson and Professor McCance emphasize that the greatest caution must be used in coming to any general conclusion on the basis of these results. The conclusions drawn must be restricted to the conditions under which the scientific evidence was obtained and cannot justifiably be applied to the needs of populations in very different states of nutrition and with widely varying dietary habits.

MEDICAL RESEARCH COUNCIL,
38, Old Queen Street,
London, S.W.1

30th September, 1954

CONTENTS

FIVE GIRLS FROM DUISBURG WHO HAD LIVED FOR A YEAR ON THE EXPERIMENTAL DIETS

From left to right:

Ursula K., aged 14 years	70% extraction flour
Ursula P., aged 9 years	100% extraction flour
Helga S., aged 12 years 10 months	85% extraction flour
Eleonore K., aged 10 years 9 months	70% extraction enriched to 85% level
Ursula B., aged 15 years	70% extraction enriched to 100% level

Studies on the Nutritive Value of Bread and on the Effect of Variations in the Extraction Rate of Flour on the Growth of Undernourished Children

PART I: THE ABILITY OF FLOURS OF VARIOUS TYPES AND EXTRACTIONS TO PROMOTE GROWTH AND HEALTH IN UNDERNOURISHED CHILDREN

Introduction

THE composition of the wartime bread was one of the preoccupations of the Medical Research Council's Accessory Food Factors Committee (1940), and it was largely due to their influence in the early part of the war that the extraction rate was stabilized at 85 per cent and that calcium was added to the flour. During the war there was an improvement in the state of nutrition of the country, and there was a confident feeling among people interested in health that the rise in the extraction rate of the flour had contributed to it. Towards the close of hostilities a conference was called by the Ministry of Food to consider the composition of the post-war loaf. The report of this conference was published in November, 1945 (Ministry of Food, 1945). At that time the medical members of the conference felt strongly that bread made from a high extraction flour was of more value in human nutrition than one made from a low extraction flour, and that it was not possible to restore all the original nutritive value by adding the known B-vitamins and iron to low extraction flours. Some members of the conference wished to see a return to white flour, improved nutritionally by enrichment, and one of the recommendations made in the report was that a flour of low extraction, suitably enriched with vitamin B_1, nicotinic acid and iron should be compared experimentally with one of higher extraction. Although not specifically stated, the members of the conference clearly intended these tests to be made on man, and a scheme was put forward for doing this at a children's home or at the National Hospital, Queen Square. Owing to various difficulties, however, nothing came of these proposals. By the end of the next year a Medical Research Council Unit was established at Wuppertal, and at a meeting of the Cereals Research Subcommittee of the Accessory Food Factors Committee held on December 30, 1946, the possibility of making such an experiment in Germany was first raised and discussed. Experimental facilities were found to be available at two orphanages and, as the result of further discussion, the scope of the experiments was somewhat enlarged. A description of them forms the basis of this report. Further details of some aspects of the work may be found in the appendices; the remainder are available and can be produced if they are required.

Experiments have also been made with rats and pigs. The diets of the animals were planned to resemble as nearly as possible those of the children taking part in these experiments.

The literature on bread is so extensive and yet in a way has so little application to the subject matter of this report that it has been decided to review it in a separate publication.

The Experimental Setting in Germany

THE ORPHANAGES

The experiments described in this report were made in 1947 and 1948 in two orphanages in Germany. One was the municipal orphanage at Duisburg, an industrial town of some 450,000 inhabitants, about 30 miles away from Wuppertal where the Unit had its headquarters and laboratories. The town had been badly bombed during the war, but the orphanage had survived. It was a rather dreary building, housing some 180 children and 30 to 40 old women. The children ranged in age from 3 to 15 years, but the 'babies' were not included in the present experiments. They took part in another investigation described by Dean (1953).

The other orphanage was in Wuppertal-Vohwinkel. The municipal orphanage in Wuppertal-Barmen had been completely destroyed during a raid on the town, and in 1947 and 1948 the children were accommodated in three smaller homes. The largest of these was the one selected for the present experiment. The building was originally a large private house, but it had been used as 'party' headquarters until the end of the war. The home was smaller than the one at Duisburg; it housed about 140 children between 4 and 15 years old.

Very few of the children in either home were orphans in the strict sense of the term. Most had one parent alive, but many were illegitimate, or their parents were in prison for such offences as stealing ration books or dealing on the 'black market'.

The orphanage at Duisburg was run by a married couple who believed in very strict discipline. The children were not allowed to speak at mealtimes, and were sent outside the room or into a corner if they did. When it was first suggested that their children should be provided with unlimited quantities of bread, the couple in charge were very thankful for they knew that the children were not getting enough to eat, but to the very end they were worried about the moral effects of allowing the children to eat as much food as they wanted. In spite of their misgivings, however, they co-operated with us and supported us in every possible way throughout the experiment.

The normal arrangement at this orphanage was for the children of 5 years and above to be split up according to age into six groups, three of boys and three of girls, and no attempt was made to keep families together. Each group occupied a living room where the children had their meals and pursued their daytime activities, and a dormitory where they slept in very crowded quarters. There was a playground where all the children spent their spare time in fine weather. Two women were in charge of each group of children, and they were responsible for them day and night.

The home at Wuppertal-Vohwinkel was in the charge of a lady whose husband had been killed during the war, and her own three children lived with the others and took part in the experiment. The children all had their meals in one large dining room, and they also had four smaller day rooms, each with its corresponding dormitory. Here, too, the normal arrangement was to divide the children up according to age and sex, but any little girls who had older sisters were allowed to be with them. Although there was no bathroom in this orphanage the children appeared to be very clean and well cared for, and the women responsible for the children took a great deal of interest in their charges, making toys for them and teaching them various handicrafts. They made the living rooms

attractive with pictures and wild flowers, and altogether the house gave one a pleasant impression. What once had been the garden of the house now formed the playground, but it was small, and the staff of the orphanage took the children into some woods a mile or so distant whenever possible.

The children in both homes attended schools in the district. About a third of them were sufficiently backward to need to go to special schools. The remainder went either to Protestant or Roman Catholic schools.

THE GERMAN RATIONS FOR CHILDREN

The German rationing system differed from the English one in that all foods were rationed. Children were classified into five divisions, 0–12 months, 1–3 years, 3–6 years, 6–10 years and 10–18 years; rations for boys and girls were the same. The allowances varied from one four-weekly rationing period to

TABLE 1

Percentage of children's calorie requirements provided by the German rations during the year before the experiments began

'Requirements' have been calculated as the average calorie intakes of well-nourished British children of the same ages (Widdowson, 1947), assuming equal numbers of boys and girls at each age.

These calorie 'requirements' were as follows:

Children ..	3–6 years	1,640 per day
Boys ..	6–10 ,,	2,130 ,, ,,
Girls ..	6–10 ,,	2,060 ,, ,,
Boys ..	10–16 ,,	2,800 ,, ,,
Girls ..	10–16 ,,	2,450 ,, ,,

Period	Percentage of requirement provided by German rations for age group:				
	3–6 years	6–10 years		10–16 years	
		Boys	Girls	Boys	Girls
May–June 1946	78	64	66	53	61
June–July 1946	78	64	66	53	61
July–August 1946	85	68	71	56	65
August–September 1946	84	78	80	63	73
September–October 1946	82	79	82	65	75
October–November 1946	91	83	86	71	82
November–December 1946	92	83	86	70	80
December 1946–January 1947	92	83	86	70	80
January–February 1947	92	83	86	70	80
February–March 1947	92	82	85	71	81
March–April 1947	92	82	85	70	80
Thereafter unchanged until the end of 1947					
Average for 12 months before the experiment began at Duisburg (June 1946–June 1947)	88	78	80	65	75
Average for 12 months before the experiment began at Vohwinkel (January 1947–January 1948)	92	82	85	70	80

Note: These figures for the German rations do not include the calories provided by Schulspeise' (see p. 4).

the next. They were at their lowest in the spring of 1946, and by August of that year they had begun to rise. Table 1 shows how the calorie value of these rations compared with the calorie intakes of well-nourished British children of similar ages before the recent war (Widdowson, 1947). The rationed calories for the children aged 3–6 years were reasonably adequate throughout, but the increases in the allowances with age were not sufficient to keep pace with the increased requirements, and it will be seen that of all the children the adolescent boys were most short of food. It was very evident from the way the older children at the orphanages cleared up every scrap of food from their plates at mealtimes that they were not getting enough to eat.

McCance and Widdowson (1951a) have pointed out that few families in Germany at this time lived solely on their rations; almost everybody acquired food in some other way. It was more difficult for institutions to get food 'off the ration' because they had no stores of goods to barter with farmers for food. On the other hand, when supplies were not sufficient to honour all the rations, shopkeepers tended to supply institutions at the expense of private families because they were better customers.

THE MEALS

The daily routine before the experiments were started was very similar in the two homes. Breakfast was served at 7.15 a.m. It consisted of bread with a smear of butter or jam, and 'Ersatz' coffee without any milk to drink. The children had to be at school at 8 a.m. and while at school during the morning they had their 'Schulspeise', which consisted of a portion of soup providing about 300 Calories. This was additional to the rationed food they received at the orphanages and has not been included in the figures given in Table 1. The midday meal, which consisted of vegetables or vegetable soup, was served in the homes at 12.30. The week's meat ration, about an ounce of cooked meat for each child, was eaten for dinner on Sunday. This meat was not always available. At Duisburg the last meal of the day was at 5.30, when the children had bread with a little butter, sausage, fish, or cheese. On 5 days of the week 'Ersatz' coffee without milk was served as the drink, but on Wednesdays and Sundays sweet soup, made from milk, water, semolina and sugar, was provided. The whole of the milk supply for the week went into these two sweet soups. The amount of milk each child received at the beginning of 1947 varied from 250 to 500 ml. a week. The older children were entitled to less than the younger ones, but they got bigger helpings of soup and therefore more milk. The milk was occasionally whole milk, but more usually skimmed.

At Vohwinkel the children were given a piece of bread and jam when they came in from afternoon school about 4 and they had their supper at 6 o'clock. This meal was very similar to the one provided at Duisburg at 5.30. At both homes the evening meal tended to be better on Sundays than on other days.

THE CHILDREN

The first impression given by the children in both homes was that they were very lively and energetic, but small for their age. Many of them looked thin, and most had pot-bellies. There was a considerable amount of skin sepsis, particularly at Duisburg, and hyperkeratosis of the upper arms was fairly common.

In order to assess the measurements of the heights and weights of the children in the orphanages at the time when they were first seen, the figures for each child

were compared with a 'normal' height and weight for a child of the same age and sex. The figures chosen as 'normal' were those of O'Brien, Girshick and Hunt (1941), who measured the heights and weights of 147,000 American children from all walks of life and living in seventeen different States; white children only were included. These workers were concerned with the construction of patterns for children's garments, and they gave the average measurements for boys and girls at yearly age intervals from 4 to 17 years. From these averages curves were constructed so that the 'normal' height and weight could be ascertained at each month of age. From the age of every boy and girl in the orphanages the height and weight of his or her American counterpart were obtained from the graphs, and the German child's height and weight were then expressed as a percentage of the 'normal' American figure. The children between 5 and 15 years old were separated into three divisions, as they were for rationing purposes, and the percentages for each division averaged. The results are shown in Table 2. On the whole the children at Vohwinkel were nearer to their 'normal' heights and weights than those at Duisburg. This may have been because these preliminary measurements at Vohwinkel were made in November, 1947, whereas those at Duisburg had been made the previous April, so that the children at Vohwinkel had had a longer time to recover from the period of greatest food shortage in 1946. The youngest age group in both homes was nearest to the 'normal', both in height and in weight. The 6–10-year old group was on the whole better in both homes than the 10–15-year old group, and the girls in both of these older age groups tended to be better than the boys.

TABLE 2

Average heights and weights of children of various ages in the two orphanages before the experiments started

The heights and weights are expressed as a percentage of the 'normal' height and weight for that age as given in the tables of O'Brien, Girshick and Hunt (1941).

Age (years) and sex	Number	Average age (years and months)	Average height as per cent of 'normal'	Number below normal height	Average weight as per cent of 'normal'	Number below normal weight
			Duisburg			
4–6 Boys+girls	16	5–5	96·5	12	96·8	11
6–10 Boys ..	47	8–0	94·3	42	90·9	39
6–10 Girls ..	27	8–0	95·9	24	92·6	22
10–15 Boys ..	38	12–0	95·5	34	89·8	33
10–15 Girls ..	41	12–4	94·6	33	90·7	30
			Vohwinkel			
4–6 Boys+girls	10	5–3	98·2	8	100·6	7
6–10 Boys ..	35	8–0	96·8	29	94·4	26
6–10 Girls ..	25	8–1	97·5	21	95·0	19
10–15 Boys ..	42	12–6	94·8	38	89·2	38
10–15 Girls ..	29	12–0	95·0	22	86·9	26

The Organization of the Experiments

The bread which was available on the German ration and which the children had been eating before the experiments started was made mainly from wheat, though other cereals were added from time to time. It was a dark brown colour, and contained more germ and bran than wholemeal bread, for part of the semolina had been removed in the milling of the wheat and was sold as a food for infants. Since one of the objects of the investigation was to study the sufficiency or otherwise of the B-vitamins in the various experimental diets it seemed unwise to have the children too plentifully supplied with these vitamins before the experiments started. For a preliminary period of 2 months, therefore, the German flour and bread in each home were replaced with flour of 70 per cent extraction or bread made from it. The allowances of bread to each child remained unaltered. The flour for making this bread was milled in England and sent to Germany for the experiments. These preliminary periods were very valuable in that they gave us an opportunity to study the running and organization of the homes, particularly their feeding arrangements, and to overcome any difficulties before the main experiments began. They also provided time for making all the preliminary examinations of the children.

After the preliminary periods, the children in each orphanage were divided into comparable groups. In one of the orphanages, at Duisburg, the limited ration of bread was then replaced by unlimited amounts of bread made from one of five different experimental flours, so that the breads in question constituted as large a proportion of the children's diets as possible. Each group was assigned a special flour. In the second home, at Wuppertal-Vohwinkel, three only of the five experimental flours were used. Here, however, the ration of bread was replaced by unlimited amounts of bread, flour, fat, sugar and jam, so that the cereals constituted only part of the additional food and therefore provided a smaller fraction of the children's total calories. The idea behind this was to test the breads at two different levels of intake. It might seem that the more calories provided by bread in a diet, the more searching the test of its nutritive value would be. This would be true if one or more of the breads contained *none* of some essential dietary constituent. If, however, even the most refined bread contained *some* of each of the essentials in question, the more of this bread in the diet, the less the likelihood of a deficiency; feeding the breads in smaller quantities might in that case be more likely to reveal nutritive deficiencies provided the calorie intake were maintained by foods which contained none of the particular dietary essentials.

All the children in both homes had the whole of the non-cereal part of their German rations, and they all had supplements of calcium and of vitamins A, D and C.

The experiments lasted for one year, from June 1947 to June 1948 at Duisburg, and from January 1948 to January 1949 at Vohwinkel. The children were tested and examined in a number of ways in order to compare the effects of the different kinds of flour upon their growth and health.

THE EXAMINATION OF THE CHILDREN

The children were kept under close observation throughout the year of the experiments. During the preliminary period, before the main experiments began, measurements of height and weight and of the girth of various parts of the body were made. Each child was examined clinically, and a sample of blood

was taken for the determination of haemoglobin, haematocrit, serum proteins and serum P-cholinesterase. Radiological examinations were made of the gastro-intestinal tract and of the bones, and the teeth were inspected.

The children were weighed and measured every fortnight while the experiments were in progress, and they were examined clinically every 3 months. At the end of the experimental year all the examinations which had been made at the beginning were repeated. Studies of the intakes and excretions of nitrogen, minerals and of the B-vitamins were also made on some of the children at intervals during the investigation.

The children's progress, as measured in all these ways, made the basis for the conclusions as to the comparative nutritive value of the different kinds of flour.

Clinical Examinations

Every child was examined before the experiments and again at 3-monthly intervals throughout the year. On each occasion the child was examined independently by two physicians (Dr. R. F. A. Dean and Dr. A. D. Barlow), whose observations were compared before the child left the room. Where any points of difference arose the child was once again examined and an agreed opinion was recorded. Neither doctor ever knew to which group a particular child belonged. As a result of these examinations the child was at once given an overall clinical grading, A, B or C, which was based on general impressions rather than on any specific attributes or shortcomings. The clinical grading at the beginning was one of the criteria used in dividing the children into the various groups.

At every clinical examination signs of specific vitamin deficiencies were looked for, but none was detected in any group at any stage of the experiments. The appearance and texture of the skin, muscular development and tone, and the amount of subcutaneous fat were also noted and graded.

Haematological and Biochemical Investigations

Blood was collected by vein puncture. One portion was taken with the minimum of constriction into a tube containing 0·02 g. of a mixture of potassium oxalate (2 parts) and ammonium oxalate (3 parts) per 10 ml. of blood. A second sample was collected without anti-coagulant and the serum was used for the determination of P-cholinesterase, albumen and total protein. The percentage of globulin was obtained by difference.

The methods used for these determinations are those described in the appendix to the Special Report (1951).

Radiological Examinations

(i) *Gastro-intestinal tracts*. Before the investigation started the gastro-intestinal tracts of 85 boys and 69 girls in the orphanage at Duisburg were examined. At the end of the experiment barium meals were given to 4 boys and 4 girls from each of the five experimental groups. Some of the children chosen from each group at the end had shown abnormal radiological appearances in their small intestines at the first examination (Berridge and Prior, 1952). Barium meals were not given to the children at Vohwinkel at the beginning, but the gastro-intestinal tracts of 2 boys from each group were examined at the end.

The children were examined in the fasting state by the following technique. They were each given 150 ml. of a suspension of 100 g. of barium sulphate in 225 ml. of normal saline, which was drunk over a period of about 5 minutes. The children were then examined fluoroscopically in the erect position, and the level of the spine to which the lowest part of the greater curvature of the stomach extended was noted. They were screened in the supine position every 30 to 45 minutes until the barium had reached the caecum. Films were exposed when it seemed desirable.

(ii) *Bones*. The left elbow and left wrist of all the children in the orphanage at Duisburg were X-rayed before the investigation began. At the end of the experiment the legs as well as the arms of all of these children who still remained in the home were examined. Radiographs were taken of the left arm and left leg of all the children at Vohwinkel at the beginning and of those who were still in the orphanage at the end of the investigation. The techniques are described in detail by Berridge and Prior on p. 119.

Dental Examinations

The children at Duisburg were inspected by an Army Dental Surgeon before the main experiment began. He recommended the extraction of a few decayed teeth which might have interfered with the mastication of large quantities of bread. Only one 15-year old girl needed dental treatment during the ensuing 17 months.

In January, 1948, 6 months after the experiment at Duisburg had started, the children's teeth were examined for structure and caries according to the procedure used by May Mellanby and her colleagues in their surveys of London schoolchildren (Mellanby, M. and Coumoulos, 1944, 1946; Mellanby, M. and Mellanby, H., 1948). The same children were re-examined 10 months later (November, 1948). The children in the orphanage at Vohwinkel were similarly examined in January and November, 1948, which coincided approximately with the beginning and end of the experimental diets.

Measurement of Heights and Weights

At Duisburg girls were weighed one week and boys the other. At Vohwinkel all the children were weighed fortnightly on the same day. Platform balances with a steelyard, weighing to the nearest 100 g., were used for the weights, and measuring rods attached to the balances and graduated to the nearest 0·2 cm. for the heights. Great care was taken to ensure that the balances and the measuring rods at the two homes agreed with each other, and that they were always correct. It was realized that the weighing and measuring of the children was one of the most important parts of the experiment, and all the measurements at both homes were always made by one of the authors (E.M.W.). The children were weighed naked, at the same time in the afternoon, and they were always sent to empty their bladders immediately beforehand. In measuring the height, gentle traction was applied to the head, and the child was made to stand as upright as he could.

As the experiments progressed, it was soon noticed that there were 'high' weeks and 'low' weeks for the measurements of body weights. On some occasions nearly all the children would weigh slightly less than they had done a fortnight before, on other occasions they would all appear to have put on more than the anticipated amount of weight. These fluctuations must have been caused by

variations in the retention of water due to some circumstances which affected all the children on the same day. They were not apparently related to the external temperature, and there was no obvious connexion between the amount of exercise the children had been taking immediately before the weighing and their weights on any particular day. It was thought that the amount of salt in the soup eaten for the midday meal 3–5 hours previously might be the cause of these variations in weight. To test this the children were weighed in the same order on two successive afternoons. On the first occasion they were given a very salt soup for their midday meal, on the second an equal amount of one containing much less salt. Out of 141 children, 91 weighed more after the salt soup, 36 less, and 14 exactly the same. The average weight was 0·16 kg. more after the salt soup than after the less salt one, and it seems as though the amount of salt taken at midday may have been one of the factors involved in these fluctuations in weight. From the practical standpoint it is important to realize that these variations may occur, because if the subjects are only weighed twice in an experiment, once at the beginning and once at the end, their growth measured as the difference between these two isolated measurements may lead to conclusions which are entirely false, particularly if the period is a short one. This has been emphasized by other workers (Corry Mann, 1926). In the present investigation the heights and weights of each individual child were plotted every fortnight, and these graphs were used to determine the points to be taken for the calculation of growth over each successive 3-month period.

Somatic Measurements

Measurements of the girth of various parts of the body were made at the beginning and end of the experimental year at both homes. The same person made the measurements on the two occasions, and they were both made at the same time of day. For all measurements a good tape-measure was used. The methods and points of reference were briefly as follows. *Circumference of upper arm*: The circumference was taken at the midpoint of the distance between the anterior tip of the acromion process to the joint line between the humerus and ulna on the external surface. *Circumference of forearm*: The greatest circumference was taken at right angles to a line pictured as running longitudinally through the centre of the forearm. Both these measurements were made on the right arm, as it hung loosely by the side. *Circumference of thigh*: The circumference was taken at the midpoint of the distance between the anterior superior spine of the iliac crest and the joint line between the femur and tibia on the inner aspect. *Circumference of calf*: The largest circumference in a horizontal plane was taken. *Outside deltoid*: The child stood upright with his arms close to his sides. The circumference was measured outside the arms at the greatest protuberance of the deltoids. *Circumference of chest*: Two measurements were taken horizontally at the highest point of the axillae, one with the chest expanded and one with the chest deflated. The mean of these two figures was used. *Iliac crests*: The horizontal circumference at the highest point of the iliac crests was taken.

THE GROUPING OF THE CHILDREN

The children at Duisburg were divided into five groups, the groups being matched as closely as possible, taking into account the age, sex, height, weight and clinical condition of each child. This was done by making for each child a small card giving his age, his clinical grading and a grading for the amount by

which he was above or below 'normal' height and weight. Boys and girls were dealt with separately. The cards for each sex were arranged in order of age, and then, starting with the oldest child and working straight through the set of cards, five cards were laid out in a row from left to right across the table, the next five were laid down below them starting from the right and working across to the left, the next five again from left to right, and so on until all the cards were used up. In this way the average ages of the five groups were equalized as nearly as possible. The clinical and the height–weight gradings in the five groups were then inspected, and interchanges were made where necessary between children of similar ages, so that each group finally contained the same number of the various clinical gradings and of the various gradings for height and weight. When there were several children belonging to one family they were put into different groups. The children at Vohwinkel were divided into three groups in a similar way.

Altogether, 169 children started the experiment at Duisburg and 141 at Vohwinkel. Every effort was made to keep the children in the homes throughout the experiments, and the authorities were very co-operative, but in spite of this many left, and details of the numbers of various ages who started and who were still present at the end of 3, 6, 9 and 12 months are given in Table 3. Table 4 shows the numbers of children in the different bread groups in the two homes who started and who completed the same periods of the experiments. During the few days between the time the children were grouped and the time

TABLE 3

Numbers of children of different ages who started and who completed 3, 6, 9 *and* 12 *months of the investigation*

Age at beginning (years)	Numbers starting		Numbers completing:							
			3 months		6 months		9 months		12 months	
	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls
					Duisburg					
4–6	6	10	6	10	6	7	4	6	3	5
6–8	25	15	25	14	22	14	19	13	18	9
8–10	22	12	20	12	20	11	17	9	14	7
10–12	17	16	16	15	16	15	14	14	13	13
12–15	21	25	21	21	20	21	16	19	9	14
Total	91	78	88	72	84	68	70	61	57	48
					Vohwinkel					
4–6	5	5	4	5	2	4	1	3	1	1
6–8	17	12	16	12	15	12	12	11	5	8
8–10	18	13	14	12	12	12	8	7	6	6
10–12	14	13	10	11	7	10	7	7	5	5
12–15	28	16	21	11	18	8	14	7	12	6
Total	82	59	65	51	54	46	42	35	29	26

TABLE 4

Numbers of children in the various bread groups who started and who completed 3, 6, 9 and 12 months of the investigation

Extraction rate of flour (per cent)	Numbers starting		Numbers completing: 3 months		6 months		9 months		12 months	
	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls
					Duisburg					
100	18	14	18	14	17	13	15	12	13	11
85	18	17	18	15	17	15	16	14	15	13
70	18	15	17	15	16	15	12	13	9	11
70 enriched to 100	18	17	17	15	16	14	13	12	9	8
70 enriched to 85	19	15	18	13	18	11	14	10	11	5
Total	91	78	88	72	84	68	70	61	57	48
					Vohwinkel					
100	27	21	20	19	17	16	14	11	9	7
70	28	19	23	16	19	14	14	13	9	10
70 enriched to 100	27	19	22	16	18	16	14	11	11	9
Total	82	59	65	51	54	46	42	35	29	26

TABLE 5

Average 'starting' ages of the children remaining in the various bread groups at different stages of the experiments

Extraction rate of flour (per cent)	Average 'starting' ages (years and months): At start		After 3 months		After 6 months		After 9 months		After 12 months	
	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls	Boys	Girls
					Duisburg					
100	9–9	9–11	9–9	9–11	9–11	10–3	9–9	10–4	9–6	10–5
85	9–5	10–1	9–4	10–1	9–7	10–1	9–5	10–2	9–5	9–10
70	9–6	10–0	9–6	10–0	9–7	10–0	9–6	9–6	9–3	10–0
70 enriched to 100	9–9	9–9	9–10	9–11	9–8	10–0	9–9	10–4	8–4	10–9
70 enriched to 85	9–5	9–9	9–3	9–9	9–3	10–5	9–8	10–8	9–6	10–7
					Vohwinkel					
100	10–1	9–9	9–9	9–5	10–3	9–3	10–7	9–6	11–1	9–3
70	10–6	9–9	10–3	9–7	9–10	9–2	9–5	9–6	10–3	10–0
70 enriched to 100	10–1	9–9	9–10	9–4	9–11	9–4	10–5	8–7	10–9	9–3

the experiment actually started at Duisburg two families of girls left the orphanage. This explains why the number of girls in the different bread groups starting the experiment varied from 14 to 17. The 'lost' children were not necessarily of similar ages in the different groups, and Table 5 shows the average 'starting' ages of the children remaining in the various bread groups at different stages of the investigations. Table 6 gives the average initial heights and weights, expressed as a percentage of the 'normal' values (see p. 5), of the children in the different groups who completed the year of the experiments.

TABLE 6

Average heights and weights at the beginning of the experiments of all children in the different groups who completed 12 months

The height and weight of each child have been expressed as a percentage of the 'normal' height and weight for that age, as given in the tables of O'Brien, Girshick and Hunt (1941).

Extraction rate of flour (per cent)	Height	Weight
	Duisburg	
100	95·8	93·4
85	95·2	93·2
70	95·8	92·7
70 enriched to 100	94·8	89·2
70 enriched to 85	95·3	93·5
	Vohwinkel	
100	97·0	95·1
70	94·7	90·0
70 enriched to 100	96·0	91·6

The losses of children have complicated the interpretation of some of the results, but the children left for reasons quite unconnected with the experiment, and there was never any suggestion that any were taken away because they were thought to be doing particularly badly or well (and see Irwin, p. 71). The losses were most serious about Easter, which was the time of year when the older boys and girls left the homes and started to work. This accounts for the big drop in numbers in the 12–15-year old children between 9 and 12 months at Duisburg, and between the beginning of the experiment and 3 months at Vohwinkel (Table 3).

THE EXPERIMENTAL FLOURS AND BREADS

All the flours used in this investigation, both for the preliminary period and the experiment proper, were milled under the direction of Dr. T. Moran, director of the Cereals Research Station, St. Albans. They were not treated with agene or any other improver. They were sent out to Germany in two batches and stored in a cool warehouse in Duisburg, from which they were drawn for use as required.

The first batch was milled from a grist consisting of 65 per cent mixed Manitoba wheats and 35 per cent English wheat. The second batch, which was milled about 6 months later, and was intended to be as like the first batch as possible, was made from a mixed grist of 57½ per cent Manitoba, 12½ per cent Hard Winter (American) and 30 per cent English wheat.

There were five different experimental flours:

1. 100 per cent extraction.
2. 85 per cent extraction.
3. 70 per cent extraction.
4. 70 per cent extraction enriched with aneurin, riboflavin, nicotinic acid and iron up to the amounts of these substances present in the 100 per cent extraction flour.
5. 70 per cent extraction enriched with the same substances up to the amounts present in the 85 per cent extraction flour.

All five flours were used at Duisburg, but only flours 1, 3 and 4 at Vohwinkel.

To avoid all chances of confusion between the enriched and unenriched 70 per cent flours and breads, the two enriched flours were coloured yellow with turmeric. No. 4 contained 0·25 and No. 5 0·1 g. per lb. All five flours contained added calcium carbonate (Creta praeparata) in the following amounts:

100 per cent extraction flour:	37 oz. $CaCO_3$ per 280 lb. flour
	or 0·33 g. Ca per 100 g. flour
85 per cent extraction flour:	17 oz. $CaCO_3$ per 280 lb. flour
	or 0·15 g. Ca per 100 g. flour
70 per cent extraction flour:	10 oz. $CaCO_3$ per 280 lb. flour
(enriched and unenriched)	or 0·09 g. Ca per 100 g. flour

The five varieties of flour were milled from each of the two grists just described. After arrival in Germany all the flours of both consignments were analysed for moisture, nitrogen, calcium, magnesium, phosphorus, iron, aneurin, riboflavin and nicotinic acid and its derivatives. To make the samples taken for analysis as representative as possible, six sacks of each flour were chosen at random from the first consignment. Five kg. of flour were weighed out from each sack, combined and carefully mixed to give a composite 30 kg. sample. In the case of the second consignment, 2 kg. from ten sacks of each flour were taken. A portion of each mixture was used for analysis. The methods used for the analyses are described by McCance, Widdowson and Shackleton (1936) and Holman (p. 92).

Table 7 shows the composition of the five experimental flours. These are average values for the two grists. The concentrations of nitrogen and inorganic constituents in the two batches were very close; variations in the B-vitamins from one batch to the other are discussed by Holman (p. 92).

The higher extraction flours contained more protein, fat, magnesium, phosphorus and iron than the white flour. In general, the concentrations of the B-vitamins in the enriched flours were similar to those in the flours they had been made to imitate. The concentration of riboflavin in the wholemeal flour as determined fluorimetrically, however, was considerably higher than that in the enriched white flour. This point also is discussed by Holman (p. 92).

The flours were analysed for fibre at the Cereals Research Station by the method of Moran and Pace (1942); the 100 per cent extraction flour contained 2·0 g. fibre per 100 g. and the 85 per cent extraction flour 0·4 g. per 100 g. The amount of fibre in the 70 per cent extraction flours was negligible.

TABLE 7

Composition of the experimental flours

Extraction rate of flour (per cent)	g. per 100 g.			mg. per 100 g.				µg. per g.	
	Moisture	Protein (N x 5·7)	Fat	Ca (including added Ca)	Mg	P	Fe	Aneurin	Riboflavin
100*	11·8	12·0	2·92	376	131	307	4·41	4·1	1·52
85	12·2	11·8	2·77	193	77·5	192	2·84	3·4	0·81
70*	12·4	10·9	1·90	109	34·0	108	1·70	1·5	0·46
70 enriched with aneurin, riboflavin, nicotinic acid and iron to 100 per cent levels*	12·4	10·9	1·90	109	35·2	108	4·20	4·1	0·98
70 enriched with aneurin, riboflavin, nicotinic acid and iron to 85 per cent levels	13·1	10·9	1·90	109	36·3	108	2·67	3·4	0·71

* Used at Vohwinkel as well as Duisburg.
† Total acid-hydrolysable derivatives.

All the vitamin values shown here were obtained by chemical methods. The results given by other met of assay are described and discussed by Holman (p.92).

OTHER FOODS SUPPLIED FOR THE EXPERIMENTS

The jam, sugar, margarine and concentrated orange juice needed for these experiments were sent out to Germany from England. The jam was South African, of various kinds, and was tinned; the margarine was the 'special' variety as sold to the public in Britain and contained 450–550 I.U. vitamin A and 90 I.U. vitamin D per oz. The sugar was granulated. The concentrated orange juice was Californian and it was diluted with 12 volumes of water before use. It was the custom in the orphanages to use semolina for thickening the milk soups. The German semolina was of unknown quality, so it was decided to supply the orphanages with sufficient semolina of low extraction to thicken the soups. The Ministry of Food arranged for the supply of this material, and it was used for children in all the bread groups.

THE DIETS

Experimental Arrangements within the Homes

Throughout these investigations the normal arrangements in the homes were interfered with as little as possible, and the regular routines were allowed to continue so long as they did not affect the experiments. Thus the children were given all the meat, fish, milk, cheese, fat, sugar and vegetables to which their German rations entitled them, and these were cooked and served in the usual way and at the usual times. All the German bread, flour, oatmeal and other cereals were, however, replaced by one or other of the experimental breads and

flours. Bread was baked from the experimental flours in German bakeries situated near the two orphanages.

At Duisburg bread was available at every meal in unlimited amounts. Some jam was provided to make the bread a little more appetizing, but it was always spread very sparingly. At Vohwinkel too the extra food was unlimited in amount, but each slice of bread was spread sufficiently thickly with margarine and jam to provide about as many calories from fat and sugar as from bread. Hence, however many calories the children took in this form, only half could be obtained from bread. It was soon found that the children ate the fat and sugar much more readily in the form of cakes and biscuits than spread on bread as margarine and jam, so biscuits were provided in the middle of the morning and cakes at teatime. The cakes and biscuits were baked without egg or milk, and baking powder was the only raising agent used. They were made from the three experimental flours used in this orphanage, and were baked by the German baker who was responsible for making the experimental breads.

As already stated, German children received a portion of soup at school. This varied considerably in ingredients and composition, and it was decided that it would be inadvisable to allow the children to have it during the course of these experiments. It was therefore arranged that the children should come back from school in the middle of the morning for a drink of orange juice with either bread and a smear of jam (at Duisburg), or biscuits (at Vohwinkel).

At Duisburg the children in each bread group ate in a different room. At Vohwinkel the children ate in one large dining room with each bread group at a separate table. The German staff who were responsible for these groups watched their children throughout the meal. The children soon developed a sense of loyalty for their particular bread, and when some of them were taken to the local Zoo with a parcel of bread scraps to feed the animals, each child instinctively picked out a piece of his or her own sort to give to the monkeys!

Throughout the experiments all the children at both homes received daily 2000 I.U. of vitamin A, 1000 I.U. of vitamin D, and 25 mg. of ascorbic acid. These vitamin supplements were given to them in the form of tablets, made by Glaxo Laboratories Limited.

The Quantitative Dietary Records

For the duration of these experiments a British dietitian and trained nurse were attached to each home, and it was their responsibility to see that nothing went wrong with the dietary side of the investigation. They supervised the German staff, ordered the right amount of bread and cakes and cut them up. The bread was cut by an electric machine into slices of known weight and spread with a known amount of jam (at Duisburg), or of jam and margarine (at Vohwinkel). The number of slices eaten by each individual child at every meal at both homes was recorded. This recording was done by the German women in charge of the children, and it was carried out with meticulous care. At Vohwinkel, the cakes, which were of known composition, were also cut into slices of known weight, and the number of slices eaten by each child recorded, as also was the number of biscuits. All other foodstuffs were given to the children in restricted amounts as their German rations and the internal economy of the orphanage allowed, and the dietitian kept a record of them every day. The only 'dishes' made from a number of ingredients were the vegetable and sweet soups. The amounts of the various ingredients in all of these soups,

and their final volumes, were recorded, so that their composition could be determined. From all these records it was possible to work out what each child ate on every day throughout the experiments. Apart from the breads and flours, which were analysed, the composition of the diets was calculated from the tables of McCance and Widdowson (1946).

The Composition of the Diets

To give a picture of the average diet of the average child in each orphanage over the year during which the nutritive value of the different kinds of flour was being investigated, the mean daily intakes of the various foods have been set out in Table 8. The age of this average child fell between 9 and 10 years, and the table also shows the amounts of the same foodstuffs eaten by British children

TABLE 8

Composition of diets in terms of foodstuffs

Foodstuff	Average amount eaten (g. per day)		
	Duisburg	Vohwinkel	Pre-war British children aged 9–10 years
Meat (cooked weight including sausage)	6	7	86
Fish (cooked weight)	11	22	24
Cheese	7	5	8
Milk, fresh (mostly skimmed)	82	113	450 (whole milk)
Butter and margarine	12	91	30 (on bread only)
Jam	53	43	24
Sugar	5	58	78
Orange juice (concentrated)	20	20	0
Potatoes	168	257	113
Root vegetables	36	38	11
Green vegetables	63	98	28
Dried pulses	2	6	4
Fruit (fresh and dried)	13	24	142
Semolina (for thickening soups) ..	34	36	127
Experimental flour			
100 per cent extraction	447	250	
or 85 per cent extraction	437	—	
or 70 per cent extraction	418	240	

of the same age before the recent war (Widdowson, 1947). The average amounts of total and animal protein, and of fat and carbohydrate in these diets are given in Table 9. The calorie intakes of the individual German children varied from 1,500 to 3,500 a day, but the overall average was between 2,000 and 2,500 in the two homes.

The basal German diet was in most respects similar in the two orphanages, and in both places by all commonly accepted standards it was inadequate. At Duisburg there was a little more cheese, at Vohwinkel a little more milk and

fish; the younger children at Duisburg received about 60 ml. and the older ones 120 ml. of skimmed milk a day. Those at Vohwinkel had 75 to 150 ml.

The amounts of bread eaten at Duisburg naturally varied with the age and appetite of the child. Those under 10 rarely ate more than 500 g. a day, while most of the big boys regularly ate 800 g. and some of them over 1,000 g. The experimental breads provided all the children with more than 70 per cent of their total calories, and those who ate 800–1,000 g. of bread a day with about 80 per cent. At Vohwinkel the children obtained 35 per cent of their calories from the experimental flours and 35 per cent from the additional fat and sugar provided for the experiment.

TABLE 9

Protein, fat and carbohydrate in the diets

Extraction rate of flour (per cent)	100	85	70 (enriched and unenriched)	100	70 (enriched and unenriched)	Pre-war British children
	Duisburg			*Vohwinkel*		
Total protein (g. per day)	73	65	61	55	51	65
Animal protein (g. per day)	8	8	9	11	11	42
Fat (g. per day)	29	26	22	93	86	92
Sugar (g. per day) ..	50	50	50	103	100	93
Starch (g. per day) ..	351	328	347	236	237	194
Percentage of total calories from protein	13·5	12·9	12·0	9·2	8·8	11·5
Percentage of total calories from fat	12·0	11·9	9·7	35·0	33·8	37·5
Percentage of total calories from carbohydrate ..	74·5	75·2	78·3	55·8	57·4	51·0

The diet at Duisburg was not low in protein (Table 9). Protein provided 12–13 per cent of the total calories, which is a slightly higher percentage than that found by Widdowson (1947) in the diets of pre-war British children who were living at home in families where there should have been no shortage of food, and where the children were eating 40–120 g. (cooked weight) of meat a day. About two thirds of the Duisburg children's protein came from the experimental bread, the remainder from the vegetables and the small amounts of meat, fish, cheese and milk. The diet at Vohwinkel was purposely lower in protein. The basal German diet there provided more than it did at Duisburg, but the amount supplied by the cereals was only about half as much. Of their total protein, the children in both homes derived only 8–11 g. a day from animal sources. They had had no more than this for the previous two years, so they could not have started these experiments with a store of those growth factors which are associated with animal foods, e.g. vitamin B_{12}.

The Duisburg diet was very low in fat. On diets containing as little fat and as much bread as this, the greater amount of fat in the higher extraction flours made an appreciable difference to the total amount of fat in the diet. It is improbable, however, that all of this extra fat can be reckoned as having been absorbed (McCance and Walsham, 1948).

Table 10 shows the average intakes of calcium, phosphorus and iron calculated from the foods listed in Table 8. The figures for iron are minimum intakes, with no allowance for iron contamination of the food from cooking pots or eating utensils. The actual intakes were probably higher.

TABLE 10

Daily intakes of calcium, phosphorus and iron on the experimental diets

Extraction rate of flour (per cent)	Daily intake (mg.)		
	Calcium	Phosphorus	Iron
	Duisburg		
100	2,320	1,890	24·1
85	1,343	1,236	14·8
70	971	863	9·8
70 enriched to 100	953	847	19·8
70 enriched to 85	981	872	13·9
	Vohwinkel		
100	1,476 + (200*)	1,181	13·5
70	844 + (200*)	723	7·2
70 enriched to 100	819 + (200*)	702	11·9

* Added to the soup as $CaCO_3$.

TABLE 11

Variation in the diet at Duisburg during the four quarters of the year

Foodstuff	Average amounts eaten (g. per day)			
	First 3 months (July-September 1947)	Second 3 months (October-December 1947)	Third 3 months (January-March 1948)	Fourth 3 months (April-June 1948)
Meat (cooked weight including sausage)	10	8	6	1
Fish (cooked weight)	3	15	13	9
Cheese	8	6	7	8
Milk, fresh (mostly skimmed)	140	55	64	68
Butter and margarine	11	10	10	14
Jam	54	49	51	52
Sugar	7	4	4	8
Orange juice (concentrated)	20	20	20	20
Potatoes	178	166	153	174
Root vegetables	23	47	53	21
Green vegetables	100	58	25	66
Dried pulses	3	4	0	1
Fruit (fresh and dried)	36	8	4	4
Semolina (for thickening soups)	38	32	34	33
Animal protein	10	8	8	6

The figures shown in Table 8 are yearly averages. The amounts of the different foodstuffs available to the children varied from one time of year to another and, since the progress of the children will be considered at different stages of the experiment, it seems worth while to record the average amounts of the foods eaten by them during each of the four quarters of the year. The values for the two orphanages are shown in Tables 11 and 12. The amounts of animal protein are also included in the tables. On the whole the variations are minor ones. In both homes the figures for milk and animal protein were highest during the late summer; this corresponded with the first 3 months at Duisburg and the third 3 months at Vohwinkel. The amount of potatoes at Vohwinkel increased with each succeeding quarter, but at Duisburg it varied very little.

TABLE 12

Variation in the diet at Vohwinkel during the four quarters of the year

Foodstuff	Average amounts eaten (g. per day)			
	First 3 months (February-April 1948)	Second 3 months (May-July 1948)	Third 3 months (August-October 1948)	Fourth 3 months (November-January 1948–9)
Meat (cooked weight including sausage)	12	7	10	4
Fish (cooked weight)	16	23	28	16
Cheese	4	6	4	4
Milk, fresh (mostly skimmed)	83	113	153	102
Butter and margarine	93	96	99	79
Jam	48	47	45	35
Sugar	58	62	57	58
Orange juice (concentrated)	20	20	20	20
Potatoes	195	214	290	320
Root vegetables	55	22	31	46
Green vegetables	75	86	116	116
Dried pulses	4	7	6	7
Fruit (fresh and dried)	19	17	30	25
Semolina (for thickening soups)	38	34	35	35
Animal protein	10	11	14	8

A Comparison of the Calorie Intakes of the Children eating the Different Kinds of Bread and Flour

Table 13 shows the daily calorie intakes of the different groups over the whole 12 months of the experiment. At Duisburg boys and girls having bread made from 100 per cent extraction flour not only ate more bread than boys and girls having flours of lower extraction, but they ate sufficiently more to make their total calorie intakes significantly higher than those of all the other children in this orphanage ($t = 2{\cdot}22$, $p = 0{\cdot}05$–$0{\cdot}02$). Table 14 shows the average calorie intakes per kg. of body weight, and again the boys and girls at Duisburg having

the 100 per cent extraction flour showed the highest figures. Such a result is in accordance with Chick's (1942) observations on rats fed on diets consisting largely of white or of wholemeal flour.

TABLE 13

Average daily calorie intakes over the whole 12 months of the experiment

Extraction rate of flour (per cent)	Average daily calorie intake			
	Duisburg		Vohwinkel	
	Boys	Girls	Boys	Girls
100	2,239	2,187	2,658	2,267
85	2,118	2,009	—	—
70	2,108	2,041	2,528	2,284
70 enriched to 100	1,976	2,108	2,489	2,191
70 enriched to 85	2,097	2,102	—	—

TABLE 14

Average daily Calories per kg. of body weight over the whole 12 months of the experiment

Extraction rate of flour (per cent)	Average daily Calories per kg. body weight			
	Duisburg		Vohwinkel	
	Boys	Girls	Boys	Girls
100	77·2	70·9	76·9	78·7
85	73·1	66·9	—	—
70	75·8	65·1	88·1	73·9
70 enriched to 100	73·3	65·9	78·9	76·2
70 enriched to 85	72·4	68·4	—	—

In order to eliminate as far as possible the effect of differences in age and size of the children in the various bread groups the results have been calculated in another way. 'Standard' tables have been drawn up, based on the results of Widdowson (1947) for the average calorie intakes of British middle-class children of various ages before the recent war. Each child's daily calorie intake over the year has been compared with these 'standards', and his intake has been calculated as a percentage of the intake of the average British child (a) of the same sex and age, and (b) of the same sex and body weight. These percentages have been averaged for each bread group, and the final means are shown in Table 15. The German children were underweight for their ages, so the percentages are lower when the German children are compared with British children of the same age than they are when compared with British children of the same body weight.

TABLE 15

Calorie intakes expressed as a percentage of the intakes of British children

Extraction rate of flour (per cent)	Total Calories compared on the basis of age	Total Calories compared on the basis of body weight
	Duisburg	
100	95·9	100·1
85	88·8	92·5
70	90·2	94·1
70 enriched to 100	89·1	95·6
70 enriched to 85	90·5	94·5
	Vohwinkel	
100	102·2	107·3
70	102·0	108·3
70 enriched to 100	99·5	104·5

Again the higher calorie intakes of the boys and girls at Duisburg having bread from flour of 100 per cent extraction are apparent. If the intakes of the boys and girls having 100 per cent and 70 per cent extraction flours are compared on the basis of age, the difference is statistically significant ($t = 2{\cdot}22$, $p = 0{\cdot}05$–$0{\cdot}02$), as it is also if the same two groups are compared on the basis of body weight ($t = 3{\cdot}38$, $p = 0{\cdot}01$).

There was no consistent difference between the total calorie intakes or the calories per kg. body weight of the three groups at Vohwinkel, probably because bread and flour contributed a very much smaller proportion of the calories in this home.

The calorie intakes of the children in all groups at Vohwinkel were higher over the course of the year than those in any group at Duisburg, whether the results are expressed as total calories, as calories per kg. body weight, or as a percentage of British children's intakes. The Vohwinkel children took about the same number of calories as British children of the same age, and about 7 per cent more than British children of the same body weight. It is striking how low all the Duisburg children's calorie intakes were, even when compared with those of British children younger than themselves but of the same body weight. Their achievements in growth are even more remarkable when this is borne in mind.

THE METABOLIC BALANCES

'Sample' children were selected from each group at both homes, and their exact intake and output of nitrogen, aneurin, riboflavin and nicotinic acid derivatives were measured over a period of one week. At Duisburg the intake and output of calcium, phosphorus and magnesium were also investigated. The metabolic studies were carried out in the orphanages during the school holidays, and the children led their ordinary lives as far as possible while they were in progress. All the studies were made on children between 11 and 15 years old.

At Duisburg 5 children were investigated at a time, one from each group, but 2 boys from each of the three groups at Vohwinkel were investigated together. These children had their meals apart from the others during the experimental week, but they were with the other children for the rest of the time. The children had been eating the experimental diets for at least 3 months before the first of the metabolic studies was made. They continued to eat exactly the same diet during the experimental week, so no preliminary period was needed before the balance experiment started.

Duplicate samples of each child's food were set aside for analysis. All urine was collected and preserved under toluene. Faeces were demarcated with carmine, and the week's collection, which was treated with HCl and toluene each day, was mixed together. The preparation of the food and the faeces for analysis and the subsequent analytical procedures for nitrogen, calcium, phosphorus and magnesium were carried out as described by McCance and Widdowson (1942) and in the appendix to the Special Report (1951). Details of the methods used for the B-vitamin assays are described by Holman (p. 92).

Results

Only those children who completed the whole 12 months of the experiments have been taken into account in presenting most of the averaged results. Tables and graphs have also been constructed for the larger number of children present for shorter periods, i.e. 6 and 9 months, but since in all instances the conclusions based upon them agree with those based on the findings for the smaller number of children over the longer period, it has been decided to present only a few of the results for the children who completed 6 or 9 months of the experiments. The numbers included in each bread group over each period may be seen in Table 4, p. 11.

Two outstanding conclusions may be drawn from these experiments. Firstly, no differences either in growth or health could be detected between the groups of children eating the various kinds of bread; secondly, *all* the children did remarkably well on these diets which by commonly accepted standards would be considered very poor. The latter point is often brought out even more clearly when all the children in each orphanage are considered together, irrespective of the kind of bread they were eating; in the presentation of the results, therefore, the progress of all the children together is considered as well as the progress of the separate groups of children eating the different breads.

CLINICAL EXAMINATIONS

General Health and Physical Condition

One of the most striking findings in these investigations, and perhaps the most unexpected one, was the remarkable way in which the general condition of all the children at Duisburg improved during the year of the experiment. Dr. J. H. Sheldon, Dr. R. E. Smith, Dr. D. M. T. Gairdner, the late Sir James Spence, and others visited Germany and saw the children towards the end of the experiments, and all agreed that they were in excellent physical shape. They were unable to pick out individual children as belonging to a particular bread group.

PLATE I

Molluscum contagiosum in a 7-year old boy at Duisburg

At no time was there any digestive upset which might have been attributed to the diet. Very few of the children had infectious diseases during the experimental year and, although there were a few cases of measles, mumps and chickenpox, there was no general spread of infection. The children recovered rapidly without any complications.

At Vohwinkel there was also a considerable improvement in general health during the year of experiment. There were some complaints at the beginning that the diet was 'sickly' because it contained too much fat, but after the fat was disguised in cakes and biscuits, nothing more was heard of the matter. Four months after the investigation had begun at this orphanage there was an outbreak of scarlet fever, which affected 6 boys and 3 girls and necessitated their being sent to hospital. These children had to be discarded from the experiment, and this was, therefore, one of the sources of loss (Table 4).

Skin Changes

When the Duisburg children were first examined many of them had infected or abnormal skins. There was a great deal of minor sepsis, particularly of the hands and feet. This latter was not surprising, because during 6 months of the year the children went barefoot or wore heavy wooden clogs without socks. Many of the children had rough dry skins, and some of them had dry scaly patches, usually on the face but sometimes on other parts of the body. Hyperkeratosis pilaris was found in 40 per cent of the children, mostly on the extensor aspects of the upper arms and thighs, which is the most common distribution in mildly undernourished persons (McCance and Barrett, 1951). A few children had scabies, which responded at once to treatment with benzoylbenzoate.

By the end of the experiment the children's skins had very much improved. There was less sepsis, for instance, due probably more to the prompt local treatment given by the nurse attached to the Unit than to the extra food. The dry scaly patches also tended to disappear and the proportion of children with hyperkeratosis fell to 16 per cent.

After the experiment had been running for about 6 months there was an outbreak of molluscum contagiosum which particularly affected the small boys (Plate I). The lesions were found in the usual sites, i.e. on the abdomen and in the axillae and antecubital fossae, and they cleared up spontaneously.

The children at Vohwinkel were better cared for than those at Duisburg and when first seen their skins were much more healthy. There was, however, some sepsis, but by the end of the experimental year its incidence was very low. Twenty-five per cent of the children had hyperkeratosis at the beginning and it was still present in 11 per cent at the end.

Muscular Development and Tone

At both orphanages only about 15 per cent of the children were recorded as having well-developed muscles when they were first seen, and 20 per cent had poor muscular development. As the experiment progressed it became increasingly evident that the children were becoming more muscular, and by the end nearly half had muscular development which was classified as 'good'; only about 7 per cent were graded as 'poor'.

Accompanying the improvement in muscular development, a similar improvement in the tone of the muscles was recorded. Some indirect evidence of the change in muscular tone is given on p. 30.

Subcutaneous Fat

During the year they were under observation the children in both homes showed an increase in the amount of their subcutaneous fat, so that by the end there were only about 7 per cent in either home whose fat was graded as 'poor'. The technique of measuring subcutaneous fat with calipers had not come into general use when these experiments were made, but there was no doubt that the children at Vohwinkel became fatter than those at Duisburg. At Duisburg, 17 out of the 105 children and 30 out of the 55 children at Vohwinkel were classified as having 'good' subcutaneous fat at the end of the investigation, and of the 30 Vohwinkel children with 'good' fat, 7 were recorded as being rather too fat.

Tendon Reflexes

Exaggerated tendon reflexes were frequently observed among the adult German population during the years of the food shortage after the war (McCance and Dean, 1951). This did not appear to be due to any physical cause, and Davis (1951) suggested that the brisk reflexes were psychological in origin and formed part of an 'effort syndrome'. Thirty per cent of the children at Duisburg had exaggerated knee jerks when first examined. By the end of the experimental year this number had fallen to 13 per cent. At Vohwinkel the initial proportion with brisk responses was 10 per cent and this figure was unchanged at the end.

The difference between the findings at the two orphanages at the beginning fits in with Davis's theory, for at Vohwinkel the children were treated with more understanding and affection than they were at Duisburg. The improvement at Duisburg may have been due at least in part to the constant presence of two members of the Unit and the frequent visits of others, which the children obviously enjoyed; the improvement in health and nutrition may also have contributed to their peace of mind, but if so, its effect, judging by the other home, can only have been small.

Signs of Puberty

It was not possible to study the sexual development of the boys taking part in the investigation, for at both orphanages they left at the age of 14 before most of them had begun to show any pubertal changes.

A comparison was made, however, of the sexual development of 26 girls at Duisburg between the ages of 11 years 6 months and 15 years 0 months at the beginning, and the same number of girls of similar age distribution at the end of the experiment. The average age on both occasions was 13 years 1 month. Sixteen of the 26 at the beginning and 17 at the end showed some breast development. At the beginning 3 girls were fully developed physically and were menstruating; at the end 6 were fully developed and had begun to menstruate. It may therefore be concluded that the girls' satisfactory growth during the experimental year was paralleled by their sexual development.

Clinical Grading

At the time when the experiments began, it was not anticipated that all the children would improve as much as they did. In fact, it was thought that some might deteriorate. The standards used for clinical gradings, therefore, were chosen in relation to the general condition of the children in the orphanage

at the first examination, so that the middle class, B, contained the largest number of children and there would be room for improvement or deterioration. Table 16 shows the numbers of children in all the bread groups at both homes classified clinically as A, B or C at the beginning and end of the experiments. Every effort had been made to maintain the same standards throughout the investigations, and, since no child deteriorated in clinical condition but many showed improvement, an unexpectedly high proportion of children achieved the grading A at the end. The children improved at both homes, and it would be impossible from these results to say that the group on any one bread did better than any other. All improved equally well as regards the special points noted at the clinical examination such as muscular development and tone, subcutaneous fat, hyperkeratosis and skin sepsis.

TABLE 16

Clinical gradings of children in the different bread groups

Extraction rate of flour (per cent)	Numbers of children classified as A, B, and C at the beginning and end of the experiment							
	Beginning			End			Total number	Number improving
	A	B	C	A	B	C		
Duisburg								
100	9	13	2	18	6	0	24	11
85	9	18	1	23	5	0	28	15
70	8	11	1	13	7	0	20	6
70 enriched to 100	4	12	1	13	4	0	17	10
70 enriched to 85	7	7	2	13	3	0	16	8
Vohwinkel								
100	7	7	2	12	4	0	16	7
70	4	14	1	14	5	0	19	11
70 enriched to 100	9	10	1	13	7	0	20	5

Table 17 shows the changes in clinical gradings when the children in each home were divided according to age and sex instead of into bread groups.

At Duisburg before the investigation began, both boys and girls over 10 years were in a better clinical state (as judged by their clinical grading) than the younger children, in spite of the fact that they were more below their standard weights. The younger boys provided most of the members of category C. By the end nearly all of the children over 10, and two thirds of the younger ones were graded A, and there were no children at all in category C. When it was realized that the original standards had been set too low, an additional grading A+ was created. It is certain that none of the children would have qualified for this grading at the beginning. Of those graded A, 7 girls and 5 boys over 10 and one girl and one boy under 10 were classified as A+ at the end. Some of these children came from each of the bread groups.

TABLE 17

Clinical gradings of boys and girls over and under 10 years

Age and sex	Numbers of children classified as A, B and C at the beginning and end of the experiment							
	Beginning			End			Total number	Number improving
	A	B	C	A	B	C		
Duisburg								
Boys over 10 ..	11	11	0	20	2	0	22	9
Boys under 10	5	25	5	23	12	0	35	23
Girls over 10 ..	16	11	0	26	1	0	27	10
Girls under 10	5	14	2	11	10	0	21	8
All children ..	37	61	7	80	25	0	105	50
Vohwinkel								
Boys over 10 ..	8	7	2	15	2	0	17	9
Boys under 10	5	7	0	7	5	0	12	2
Girls over 10 ..	4	6	1	8	3	0	11	5
Girls under 10	3	11	1	9	6	0	15	7
All children ..	20	31	4	39	16	0	55	23

At Vohwinkel the difference between the older and younger children at the beginning was not so evident. By the end, as at Duisburg, most of the older children and nearly two thirds of the younger ones were graded A and none was graded C. Of those classified as A at the end, 4 girls and 2 boys over 10 were graded A+. No child under 10 reached this class.

Thyroid Enlargement

As part of the 3-monthly clinical examinations the size of the thyroid gland was noted and graded as (a) normal (not enlarged), (b) slightly enlarged or (c) conspicuously enlarged. When the children were first examined, 5 at Duisburg and 3 at Vohwinkel were found to have conspicuously enlarged thyroids (Table 18). After the experiment had been running for 6 months at Duisburg it was noticed that some of the older girls who had had normal glands at the beginning now had glands which were definitely large, and there was also a general increase in the incidence of slight enlargement. It was thought that this might be because the supply of iodine in the diet had been inadequate for the rapid growth which had occurred during the preceding 6 months. Accordingly, supplementary iodine as potassium iodide was given to all the children, so that the older girls received an extra 30 μg. a day and all the other children 3 μg. a day. In spite of this, during the following 6 months, although the growth rate was less rapid and the children were receiving the supplementary iodine, the thyroids continued to enlarge, and by the end of the year 22 children had conspicuously enlarged glands and 42 had glands which were slightly enlarged. The 34 children who remained under observation without any alteration in their diet for yet another 6 months (p. 43) showed no further change in the size of their thyroids.

The experiment at Vohwinkel started at the time when the increasing thyroid enlargement was first noticed at Duisburg, so the Vohwinkel children were

TABLE 18

Thyroid enlargement

Age and sex	Total no. of children	Number of children showing thyroid enlargement during the experiment					
		Beginning		After 6 months		After 1 year	
		Slight enlargement	Conspicuous enlargement	Slight enlargement	Conspicuous enlargement	Slight enlargement	Conspicuous enlargement
Duisburg							
Boys over 10 ..	22	8	1	5	1	9	6
Boys under 10	33	1	1	6	1	16	3
Girls over 10 ..	23	3	3	4	9	6	11
Girls under 10	20	0	0	1	0	11	2
All children* ..	98	12	5	16	11	42	22

* The findings for 7 children who missed the 6-monthly examination have been omitted all through the table.

Age and sex	Total no. of children	Beginning		After 1 year	
		Slight enlargement	Conspicuous enlargement	Slight enlargement	Conspicuous enlargement
Vohwinkel					
Boys over 10 ..	17	6	0	4	2
Boys under 10	12	0	0	5	0
Girls over 10 ..	11	1	1	6	1
Girls under 10	15	2	2	8	1
All children ..	55	9	3	23	4

given an iodine supplement from the start. Nevertheless, in this home also, there was an increase in the number of slightly enlarged glands during the year, but only two became conspicuously large and one got smaller. In neither home was there any evidence that the kind of bread eaten had any effect upon thyroid enlargement.

It is now realized that the dose of iodine which was given was extremely small (see Murray, Ryle, Simpson and Wilson, 1948); and it did not, in fact, prevent a further increase in the size of the glands. The interesting point, however, is why they should have started to enlarge at all. There is evidence that a relationship exists between the level of calcium intake and the incidence of goitre and that an increased calcium intake in some way enhances the goitre-producing effect of any given diet (Murray *et al.*, 1948). The children's calcium intakes undoubtedly increased at the start of the experiments, and it may be that their iodine intake was just sufficient to prevent enlarged thyroid glands at a low level of calcium intake, but that it was insufficient when the calcium intake was increased.

HAEMATOLOGICAL AND BIOCHEMICAL INVESTIGATIONS

Table 19 shows the average concentrations of total protein, of albumen and of globulin in the serum of the children eating the different kinds of bread.

TABLE 19

Concentration of the serum proteins

Extraction rate of flour (per cent)	Serum protein (g. per 100 ml.)					
	Total protein		Albumen		Globulin	
	Beginning	End	Beginning	End	Beginning	End
Duisburg						
100	7·27	7·19	4·66	4·72	2·61	2·47
85	7·23	7·13	4·77	4·70	2·46	2·43
70	7·08	7·28	4·59	4·71	2·49	2·57
70 enriched to 100	7·33	7·28	4·70	4·79	2·63	2·59
70 enriched to 85	7·25	7·37	4·65	4·71	2·60	2·66
Vohwinkel						
100	7·27	7·13	4·86	4·74	2·41	2·39
70	7·20	7·11	4·72	4·65	2·48	2·46
70 enriched to 100	7·16	7·05	4·70	4·72	2·46	2·33

TABLE 20

Haemoglobin, haematocrit and serum P-cholinesterase levels

Extraction rate of flour (per cent)	Haemoglobin (g. per 100 ml.)		Haematocrit (per cent)		Serum P-cholinesterase (c.mm. CO_2/ml./min.)	
	Beginning	End	Beginning	End	Beginning	End
Duisburg						
100	12·6	12·8	40·5	40·4	75	80
85	12·9	13·1	41·2	41·2	75	78
70	12·8	13·1	41·4	41·3	71	78
70 enriched to 100	12·7	13·0	40·7	40·8	76	79
70 enriched to 85	12·5	12·9	39·9	40·2	73	80
Vohwinkel						
100	13·0	12·6	42·0	41·7	78	82
70	12·8	12·5	41·4	41·2	79	82
70 enriched to 100	12·8	12·5	41·6	40·7	82	87

There was no appreciable change in any group at either home. This result might have been expected from the fact that all the values were normal at the beginning.

Before the experiments began the levels of haemoglobin and haematocrit (Table 20) were lower than the values which are generally considered normal for adults, and they were also lower than values which have been reported for British and Australian children of similar ages (Medical Research Council's Committee on Haemoglobin Surveys, 1945; Fysh, 1950). There was no significant rise either in haemoglobin or in haematocrit in any group, in spite of the fact that the iron intakes of the children in some groups were more than twice those of children in others (Table 10, p. 18).

In children of all bread groups and all age groups at both homes the P-cholinesterase activity of the serum was higher at the end than at the beginning of the period of unlimited food (Table 20). This is in accordance with previous observations on adults (Hutchinson, McCance and Widdowson, 1951). The activity, however, did not reach the level displayed by British children of similar ages (Hutchinson and Widdowson, 1952). The averaged results for children of different ages, irrespective of the kind of bread they ate, are set out graphically in Fig. 1. The increase is another reflection of the improved

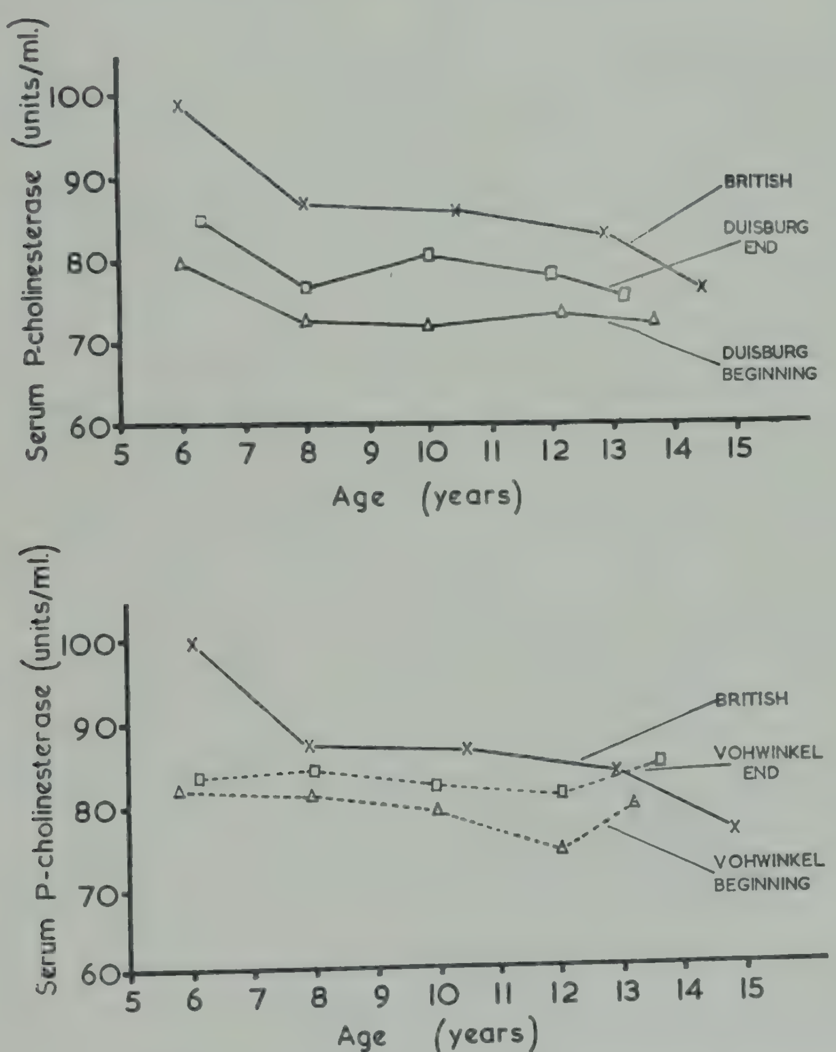

FIG. 1. Serum P-cholinesterase activity in children of different ages at the beginning and end of the experiment.

nutritional status of the German children, and the failure to reach the standards of the British children is an indication that even at the end of the experiment the German children still had some leeway to make up. A similar conclusion may be drawn from a consideration of the heights and weights (p. 36).

RADIOLOGICAL EXAMINATION OF THE GASTRO-INTESTINAL TRACTS

The gastro-intestinal tracts of 154 children at Duisburg were examined before the experiment began. In every case the shape of the stomach was normal. At the end of the investigation barium meals were given to 40 of these children, 4 boys and 4 girls from each bread group. In 11 boys and 12 girls, distributed over all the bread groups, the level of the greater curvature of the stomach was a half to one and a half vertebral bodies higher on the second occasion. This was presumably due to the increased tone of the abdominal musculature and an increase in the retroperitoneal fat. These findings resemble those previously reported in 19 undernourished men who were given unlimited amounts of food for 8 weeks (Berridge, 1951). In 4 boys and 1 girl the greater curvature of the stomach was a little lower at the second examination.

At the beginning the gastric emptying times (mean $1\frac{1}{2}$ hours, S.D. $\frac{1}{2}$ hour) and the small intestinal transit times (mean $2\frac{1}{2}$ hours, S.D. 1 hour) of the 154 children were within normal limits. The calibre and the mucosal folds of the small gut were normal.

There was no significant difference between the mean gastric emptying times or the mean small intestinal transit times of the children in the different bread groups at the end of the year of the experimental diets; nor had the mean gastric emptying times or the mean small intestinal transit times changed as a result of the experimental diets. No differences were observed between the calibre of the gut or the character of the mucosal folds of the children in any of the five bread groups.

The small intestines of 22 boys and 27 girls out of a total of 85 boys and 69 girls showed the radiological abnormalities of segmentation and flocculation before the experiment started. The significance of these findings has been discussed elsewhere (Berridge and Prior, 1952). Twenty of the 40 children chosen for the radiological examinations at the end of the experiment had shown these abnormal radiological appearances in their small intestines at their first barium meals. Similar appearances were observed in most of the same children at the end.

At the end of the year of experimental diet at Vohwinkel the gastric emptying times of the 2 boys from each of the three bread groups did not differ significantly from those of the Duisburg children, but their small intestinal transit times (mean $4\frac{1}{2}$ hours, S.D. $1\frac{1}{4}$ hours) were significantly slower ($t = 2{\cdot}2$, $p = 0{\cdot}05$–$0{\cdot}02$) than those of 12 boys of similar ages in the Duisburg orphanage after the year of experimental feeding (mean $2\frac{1}{2}$ hours, S.D. $1\frac{1}{2}$ hours).

The results of the radiological examinations of the gastro-intestinal tracts made at both orphanages have been given in detail by Berridge and Prior (1952).

DENTAL CARIES

During the experimental period in both orphanages the increase in caries was trifling in all the bread groups, and it may be concluded that the experimental diets did not affect the teeth adversely. The dietary additions at Vohwinkel, which included refined sugar and jam, would have been expected by many

people to have increased the prevalence of caries. Any harmful effect due to the sugar may have been offset by the supplements of vitamin D and calcium which all the children received.

The findings are described and discussed in detail by Mellanby (p. 131).

HEIGHTS AND WEIGHTS

It has been thought desirable to consider the heights and weights of the children in several different ways owing to the fact that the losses of children throughout the year upset the balance of the original groups.

Changes over Twelve Months

Figs. 2 and 3 show the average gains in height and weight of the five groups of children who completed the year of the experiment at Duisburg, and Figs. 4 and 5 give the same information about the three groups at Vohwinkel. At both homes the curves for height for the different bread groups are almost superimposed. There are small differences in the curves for weight, the children at Duisburg who were eating the unenriched white bread gaining rather more weight than those eating bread made from flour of 85 or 100 per cent extraction; at Vohwinkel the children having the wholemeal flour gained most weight, and those eating the unenriched white flour the least.

Since the groups were not exactly comparable as regards age and sex, the average gains to be expected of the children in them, i.e. the 'normal' gains, were not necessarily the same in all the groups. Thus, the group at Duisburg having white bread should have gained an average of about 0·2 kg. more during

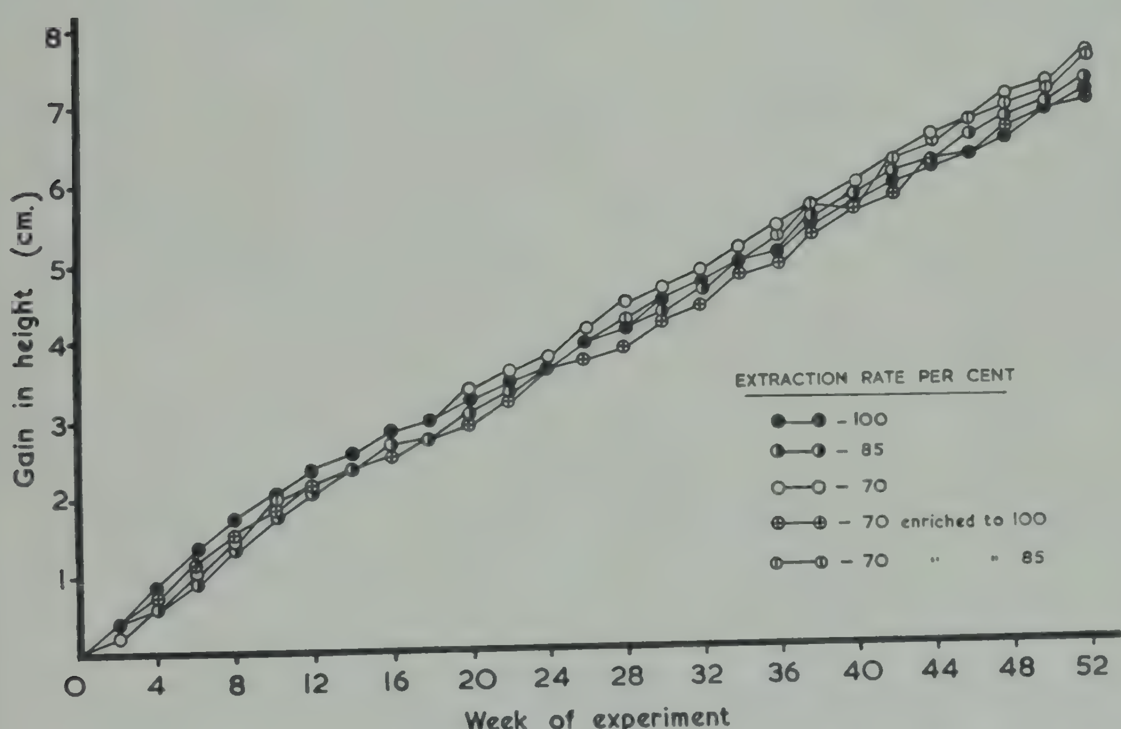

FIG. 2. Average gain in height of five groups of children at Duisburg who completed the year on the experimental diets.

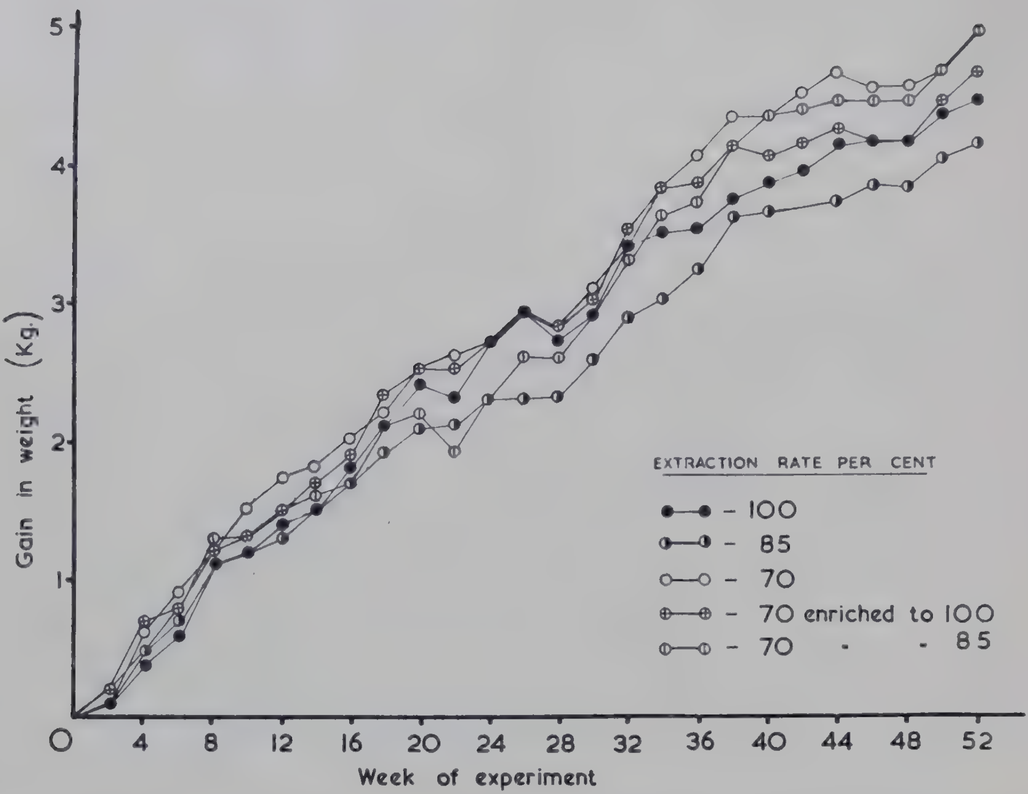

FIG. 3. Average gain in weight of five groups of children at Duisburg who completed the year on the experimental diets.

the year than the group having wholemeal bread. At Vohwinkel the children having 100 per cent extraction flour should have gained 0·6 kg. more than those having white flour. It will be seen that the actual differences were of the same order as these amounts.

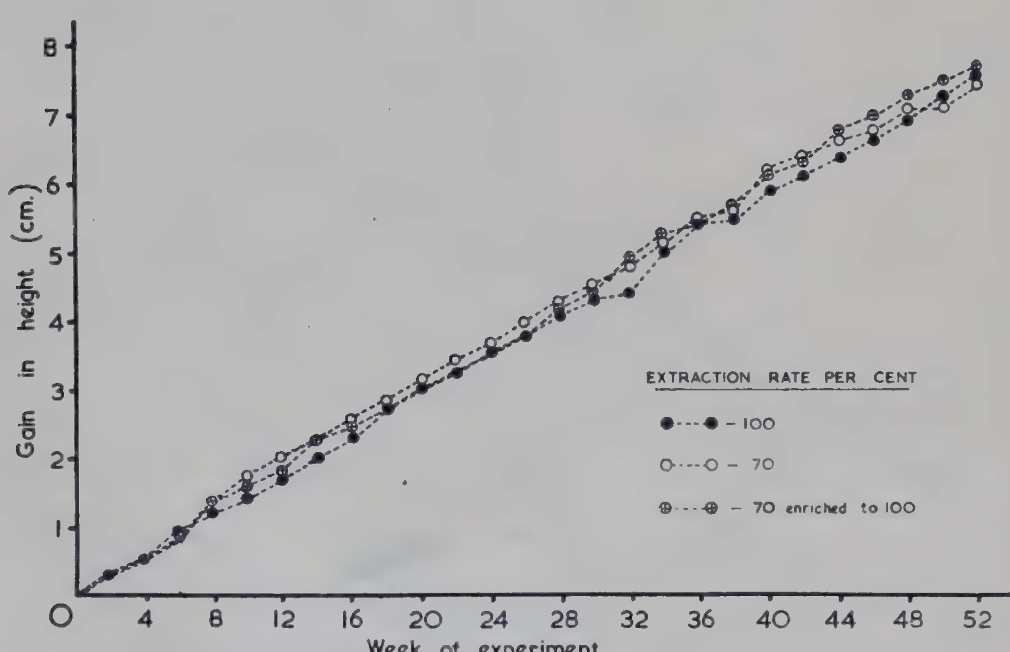

FIG. 4. Average gain in height of three groups of children at Vohwinkel who completed the year on the experimental diets.

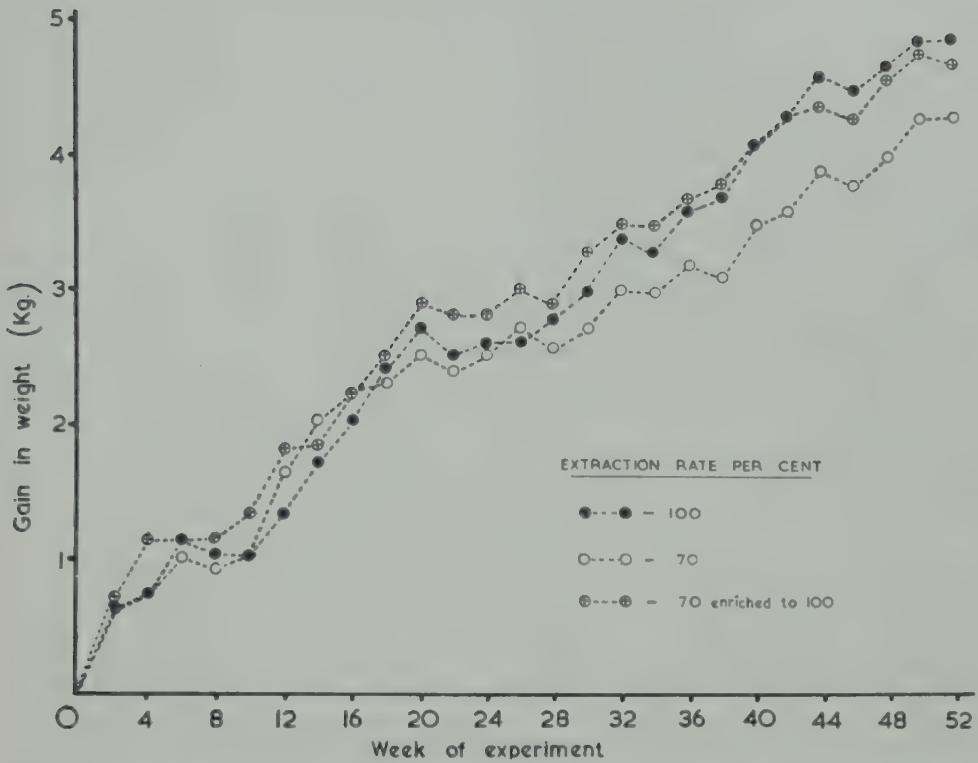

FIG. 5. Average gain in weight of three groups of children at Vohwinkel who completed the year on the experimental diets.

Changes over Six Months

Fig. 6 (p. 34) shows the average gains in height and weight of all the children in the five bread groups who completed 6 months of the experiment at Duisburg, and Fig. 7 (p. 34) gives the same information about the three groups at Vohwinkel. The number of children in each group is given in Table 4 (p. 11); a total of 152 children took part in the experiment for 6 months at Duisburg and 100 at Vohwinkel, whereas only 105 at the former and 55 at the latter home completed the full year.

The general conclusions to be drawn from these graphs are the same as those already drawn from Figs. 2–5. There was no consistent difference between the average growth rates of any of the groups.

Gains as a Percentage of the Heights and Weights at the Beginning

Table 21 shows the average heights and weights of the children at Duisburg at the beginning, and the gains over the year expressed as a percentage of the starting values. The figures for boys and girls are given separately. The group eating unenriched white flour did as well as the other groups, and slightly better on the whole than the groups having flours of 100 or 85 per cent extraction. Those who did best were boys and girls in the group having 70 per cent flour enriched to 85 per cent levels. All these differences were probably due to chance.

At Vohwinkel, as at Duisburg, there was no consistent difference between any of the groups (Table 22). For example, the boys having the 70 per cent extraction flour gained least height and weight but the girls in that group gained the most.

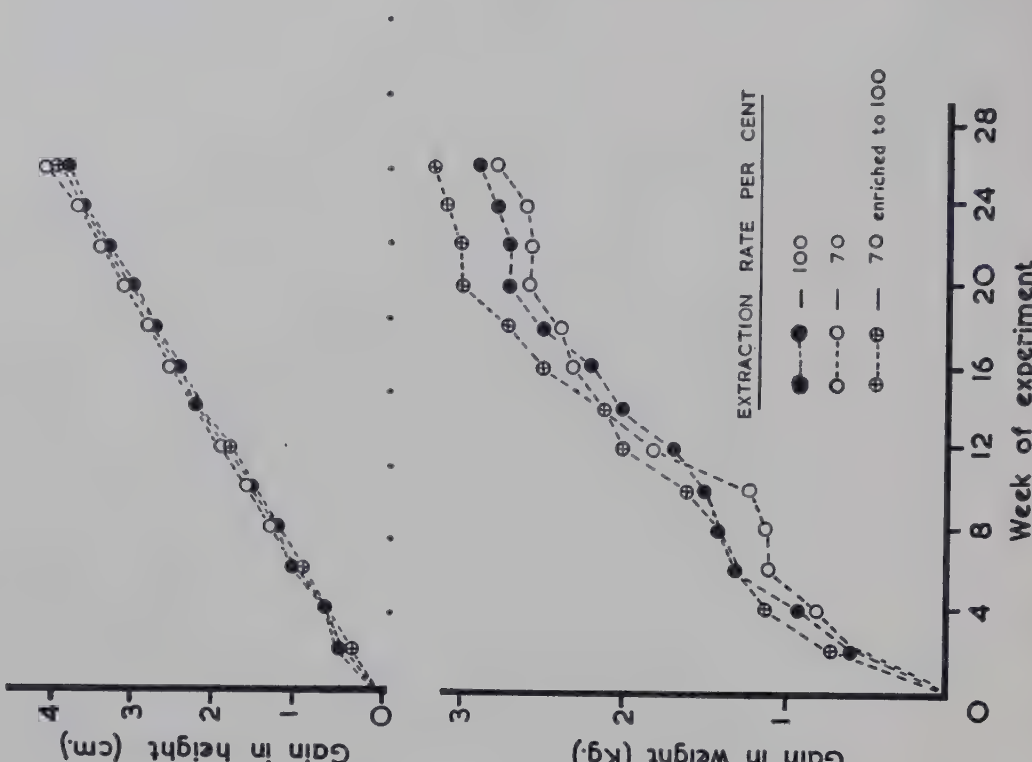

FIG. 7. Average gains in height and weight of three groups of children who completed 6 months of the experiment at Vohwinkel.

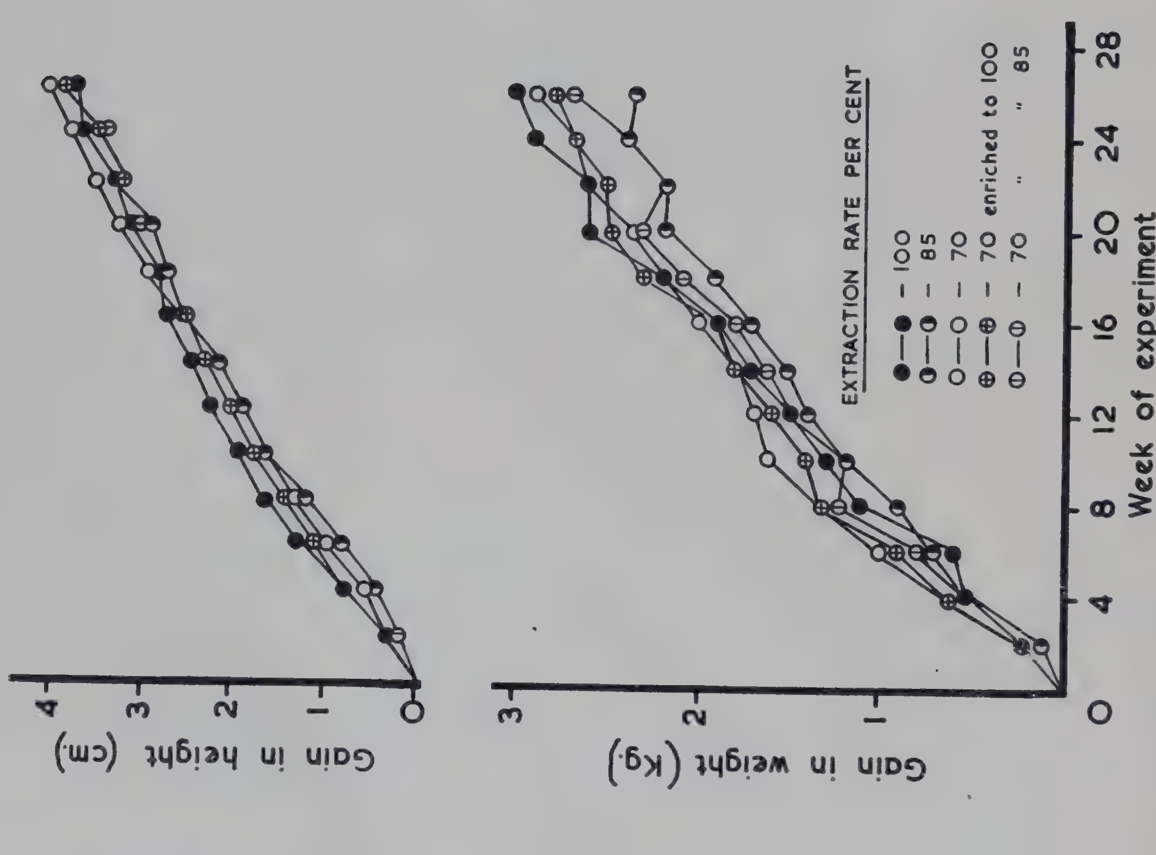

FIG. 6. Average gains in height and weight of five groups of children who completed 6 months of the experiment at Duisburg.

TABLE 21

Growth in height and weight of the children in the different bread groups at Duisburg during the 12 months of the experiment

Extraction rate of flour (per cent)	Average height at beginning (cm.)		Gain as percentage of height at beginning	
	Boys	*Girls*	*Boys*	*Girls*
100	128·4	129·1	5·3	5·7
85	126·2	128·3	6·1	5·3
70	125·1	131·4	5·8	5·9
70 enriched to 100	120·9	132·4	5·5	5·6
70 enriched to 85	126·9	127·3	5·8	6·0
	Average weight at beginning (kg.)		Gain as percentage of weight at beginning	
	Boys	*Girls*	*Boys*	*Girls*
100	27·5	29·5	12·7	18·0
85	27·2	28·1	14·7	14·9
70	26·5	30·3	15·8	18·5
70 enriched to 100	23·9	29·4	15·5	19·0
70 enriched to 85	27·4	28·5	16·4	20·7

TABLE 22

Growth in height and weight of the children in the different bread groups at Vohwinkel during the 12 months of the experiment

Extraction rate of flour (per cent)	Average height at beginning (cm.)		Gain as percentage of height at beginning	
	Boys	*Girls*	*Boys*	*Girls*
100	136·8	128·1	5·6	5·7
70	128·2	129·7	5·4	6·0
70 enriched to 100	133·4	126·7	6·1	5·7
	Average weight at beginning (kg.)		Gain as percentage of weight at beginning	
	Boys	*Girls*	*Boys*	*Girls*
100	32·6	26·5	16·9	15·8
70	27·9	29·0	12·9	16·9
70 enriched to 100	30·1	26·8	16·9	15·7

Comparison with 'Normal' Children

Figs. 8 and 9 show the average heights and weights at fortnightly intervals of all the children who completed the year of the experiments at Duisburg and Vohwinkel respectively. The heights and weights of 'normal' American children of the same ages (O'Brien, Girshick and Hunt, 1941) are indicated, and the amounts by which the German children were below these 'normal' values are shown by arrows. In both orphanages the children were under-height and underweight before the experiments began. They gained more height and weight than 'normal' children would have been expected to do in the course of a year, so that by the end of the experiment they were nearer the 'normal' height and weight for their age than they had been at the beginning. They were still, however, not quite up to 'normal' standards.

The gains in height and weight of the children in the different bread groups have been expressed by comparing them with the 'normal' rates of increase of the American children. Table 23 shows the ratio of the actual increase to the 'normal' increase of the children in both homes during the year of the experiment. Again the children having the unenriched white flour did quite as well as the other children, and certainly as well as those having the wholemeal flour. The children in all the groups gained more than the 'normal' amounts.

TABLE 23

Growth in height and weight of the children in the different bread groups expressed as the ratio of the actual increase to the 'normal' increase (*p.* 5)

Extraction rate of flour (per cent)	Height	Weight
Duisburg		
100	1·42	1·47
85	1·34	1·28
70	1·48	1·51
70 enriched to 100	1·47	1·55
70 enriched to 85	1·43	1·61
Vohwinkel		
100	1·37	1·40
70	1·33	1·47
70 enriched to 100	1·44	1·34

Boys and girls under 10 and over 10 years at both orphanages gained considerably more height and weight during the year than was to be expected of them from the 'normal' growth curves compiled from O'Brien, Girshick and Hunt's (1941) tables (Table 24). The girls tended to do better than the boys, although they were nearer to their 'normal' heights and weights at the beginning.

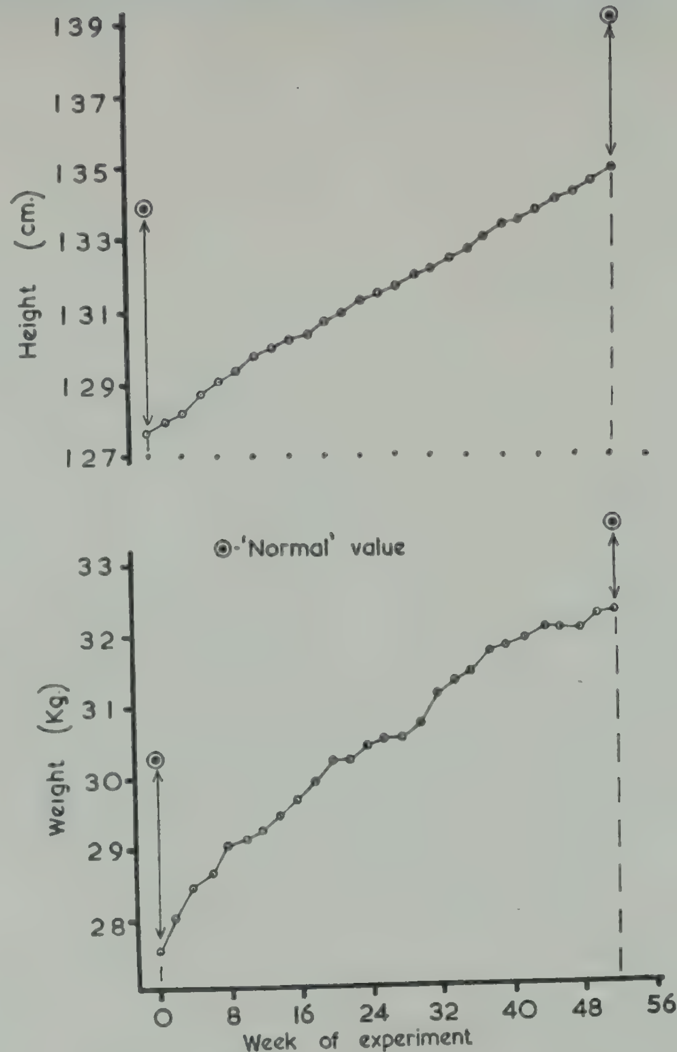

FIG. 8. The average heights and weights at fortnightly intervals of all the children who completed the year of the experiments at Duisburg. The heights and weights of similar groups of 'normal' children (p. 5) are indicated, and the amounts by which the German children were below these 'normal' values are shown by arrows.

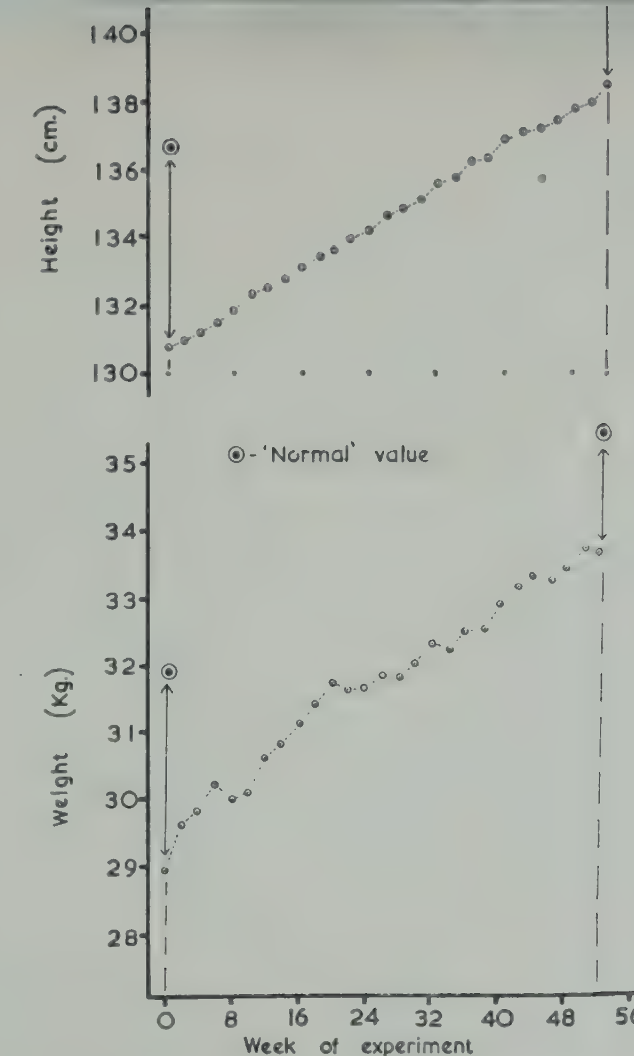

FIG. 9. The average heights and weights at fortnightly intervals of all the children who completed the year of the experiments at Vohwinkel. The heights and weights of similar groups of 'normal' children (p. 5) are indicated, and the amounts by which the German children were below these 'normal' values are shown by arrows.

TABLE 24

Growth in height and weight of children above and below 10 years, irrespective of the kind of bread eaten, compared with the 'normal' growth

Results expressed as the ratio of actual increase/'normal' increase

Age and sex	Height	Weight
Duisburg		
Boys over 10 ..	1·35	1·20
Boys under 10 ..	1·39	1·44
Girls over 10 ..	1·52	1·59
Girls under 10 ..	1·40	1·49
Vohwinkel		
Boys over 10 ..	1·33	1·21
Boys under 10 ..	1·46	1·39
Girls over 10 ..	1·56	1·39
Girls under 10 ..	1·43	1·48

Gains in Weight per 1,000 Calories and per Gramme of Protein

The children at Duisburg having wholemeal bread always tended to eat enough food to provide themselves with more calories than those eating any other kind of bread, and the wholemeal bread contained rather more protein than the others. If, therefore, the gain in weight is expressed per 1,000 Calories or per g. of protein eaten (Tables 25 and 26) the children having the brown flour showed the smallest gains in weight. Those eating bread made from flour of 85 per cent extraction gained amounts intermediate between the gains of the children having the wholemeal and the white flours. There was no consistent difference between the total calorie intakes of the three groups at Vohwinkel (p. 20), and the gains in weight per 1,000 Calories or per g. of protein showed less variation from one group to another.

TABLE 25

Average gain in weight per 1000 Calories

Extraction rate of flour (per cent)	Duisburg (g.)	Vohwinkel (g.)
100	5·31	5·35
85	5·48	—
70	6·46	4·88
70 enriched to 100	6·20	5·35
70 enriched to 85	6·23	—
All boys	5·12	5·01
All girls	6·73	5·39

TABLE 26

Average gain in weight per g. of protein eaten

Extraction rate of flour (per cent)	Duisburg (g.)	Vohwinkel (g.)
100	0·16	0·26
85	0·17	—
70	0·22	0·25
70 enriched to 100	0·21	0·28
70 enriched to 85	0·21	—
All boys	0·17	0·25
All girls	0·22	0·28

The gain in weight per 1,000 Calories was greater at Duisburg than at Vohwinkel, but the gain in weight per g. of protein was greater at Vohwinkel. These results suggest that the Duisburg diet was the more economical from the point of view of calories, while the Vohwinkel diet was the more economical from the point of view of protein. Thus there was a difference in the children's response to the diets at the two homes. In both homes the girls gained more weight than the boys per unit of protein or of calories.

Relation between Starting Weight, Gain in Weight and Calorie Intake

Each child's weight at the beginning of the experiments, his weight increase over the year and his average daily calorie intake over the year have been compared with the 'normal' values for a child of the same age and sex (see pp. 5, 20 for standards used). The results for each child have been expressed as a percentage of the 'normal' value. Scatter diagrams have been constructed relating each of these three percentages to the other two and, taking all the children together regardless of the kind of bread they were eating, regression lines have been drawn through the points (Fig. 10).

Fig. 10A shows that (a) the more a child weighed as a percentage of the 'normal' for his age at the beginning, the more he ate during the experiment; and (b) for any given percentage over- or under-weight, the children at Vohwinkel ate more in terms of calories than those at Duisburg. The heavier children showed this most. Fig. 10B shows that (a) the more a child weighed as a percentage of the 'normal' for his age at the beginning, the more weight he gained in proportion to the 'normal' increase, and (b) the Duisburg children gained more weight for a given percentage underweight than the Vohwinkel children. This difference was not so marked for the overweight children. Fig. 10C, which relates calorie intake to growth in weight, shows that the more a child ate in proportion to the standard the more weight he gained.

Thus it may be concluded that the heavier a child was for his age at the beginning of the experiment the more he ate and the more he grew. The explanation for this is no doubt that in a mildly undernourished community the most underweight are on the whole those who are by nature thin (McCance and

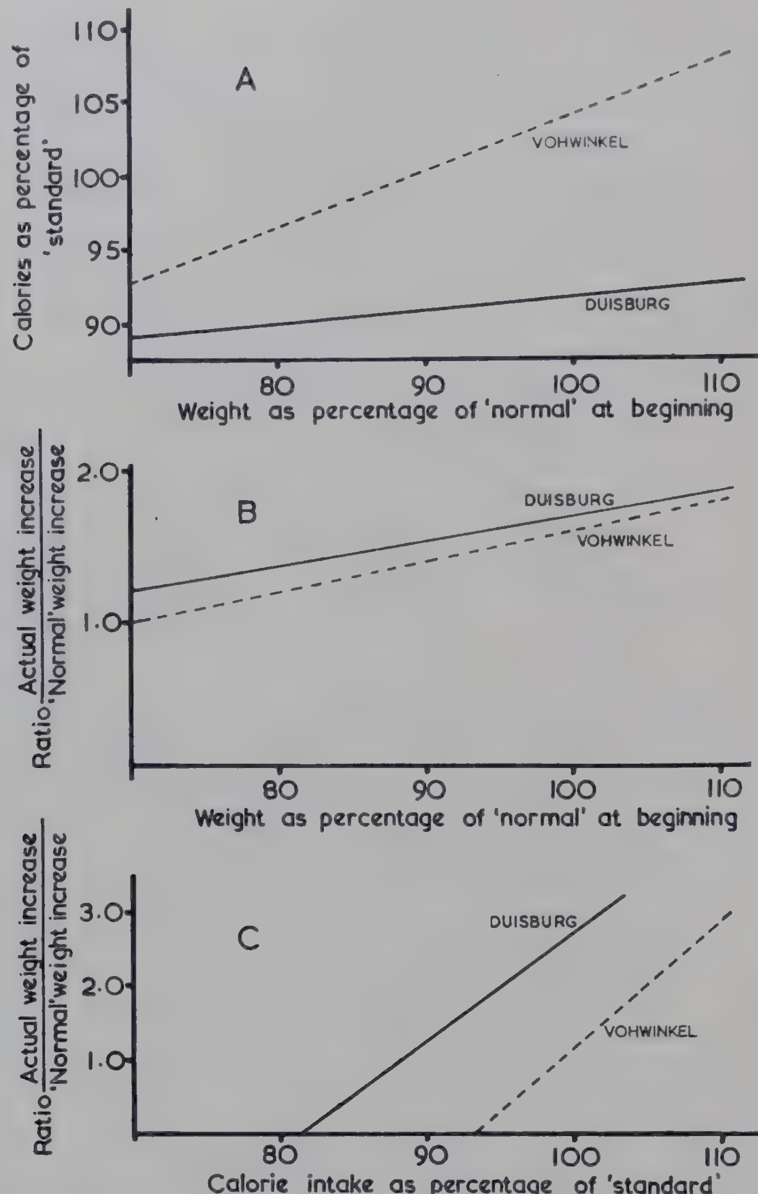

FIG. 10. The relation between calorie intake, weight at beginning and gain in weight.

Widdowson, 1951b). These persons would never eat enough to get fat. Those who are the heaviest when they are undernourished are those who would also be the heaviest when they are well fed.

Bone Development and Growth

The children's bones were under-developed for their ages at the beginning of the experiments (Table 27) and during the year they matured, as the height and weight increased, with more than normal rapidity (Berridge and Prior, p. 119). The diets, therefore, appeared to have been satisfactory in this respect as in other ways.

There was no significant difference between the bread groups as judged by bone development during the year of the experiments (Table 28). Hence it would appear that the progress of ossification had not been influenced by the extraction rate of the flour at either orphanage. All the groups gained more in this respect than one would have expected in 12 months.

TABLE 27

A comparison of the development of the children's bones with growth in height and weight during the year of the experiments

Age and sex	Months behind in age deduced from bone development of hand		Months behind in age deduced from the 'normal' height curves		Months behind in age deduced from the 'normal' weight curves	
	Beginning	End	Beginning	End	Beginning	End
Duisburg						
Boys over 10 . .	15	6	12	7	8	5
Boys under 10 . .	22	16	13	9	9	4
Girls over 10 . .	16	11	14	9	8	3 ahead
Girls under 10 . .	16	11	10	5	7	1 ahead
Vohwinkel						
Boys over 10 . .	17	11	17	12	14	10
Boys under 10 . .	28	21	10	5	7	2
Girls over 10 . .	27	18	17	13	15	9
Girls under 10 . .	17	12	6	1 ahead	1	4 ahead

TABLE 28

The mean growth in age of each bread group assessed from the development of the bones of the children's hands during the year of the experiments

(see Berridge and Prior, p. 119)

Extraction rate of flour (per cent)	Duisburg			Vohwinkel		
	Mean years	*Mean* months	Standard deviation months	*Mean* years	*Mean* months	Standard deviation months
100	1	6	± 6	1	7	± 6
85	1	6	± 5	—	—	—
70	1	5	± 5	1	5	± 4
70 enriched to 100	1	5	± 5	1	6	± 5
70 enriched to 85	1	6	± 5	—	—	—

Changes in each Successive Three Months

At neither home was the gain in height and weight in the first 3 months maintained throughout the year (Table 29). At Vohwinkel the gain in weight was less in each succeeding 3-month period, but at Duisburg the increments in both height and weight were greater during the third quarter than during the second. In the fourth quarter the gains in weight at Duisburg were relatively small and were, in fact, less than the 'normal' increase for a group of children of these ages; at Vohwinkel the actual gain in weight during the fourth quarter

TABLE 29

Gain in height and weight over the four quarters of the year of the experiments

	Gain in height and weight during:			
	1st 3 months	2nd 3 months	3rd 3 months	4th 3 months
Duisburg: height (cm.)	2·39	1·59	1·78	1·50
Vohwinkel: height (cm.)	2·03	1·91	1·97	1·63
Duisburg: weight (kg.)	1·55	1·10	1·33	0·55
Vohwinkel: weight (kg.)	1·75	1·06	0·93	0·88

was approximately equal to the 'normal' gain. At both homes the gains in height during the last quarter continued to be greater than the 'normal' gains.

At Duisburg the children ate approximately the same amount all through the year (Table 30), and the greater gains in height and weight at the beginning could not be explained by a greater intake of calories at that time. The children

TABLE 30

Average daily calorie intakes during the four quarters of the year of the experiments

Home	Average daily calorie intake during:			
	1st 3 months	2nd 3 months	3rd 3 months	4th 3 months
Duisburg	2,087	2,127	2,120	2,074
Vohwinkel	2,503	2,458	2,357	2,317

at Vohwinkel, on the other hand, ate less and less as the experiment proceeded, so that in the fourth quarter, when they were nearly a year older, the Calories were about 200 a day less than they were in the first quarter. The decreasing weight gains at Vohwinkel were therefore paralleled by decreasing calorie intakes.

The falling off in calories at Vohwinkel was most apparent during the first few weeks after the children had started to eat the experimental diets. The average daily intakes were 3,175, 2,852, 2,580 and 2,491 during the first four experimental weeks.

At both homes the children gained much less weight for every 1,000 Calories at the end than at the beginning (Table 31), although at Vohwinkel the gain in weight per 1,000 Calories fell off less in the course of the experiment than the absolute gain.

Clearly the outstanding question to be answered is: To what was the poorer growth at the end of the experiments due? Was it a reflection of the fact that the children were approaching their 'normal' heights and weights, and therefore

TABLE 31

Gain in weight per 1,000 Calories during the four quarters of the year of the experiments

Home	Gain in weight (g.) per 1,000 Calories during:			
	1st 3 months	2nd 3 months	3rd 3 months	4th 3 months
Duisburg	8·19	5·71	6·88	2·94
Vohwinkel	7·66	4·72	4·34	4·14

had little or no leeway to make up, or was it due to some sort of dietary deficiency?

The children in all bread groups at Duisburg gained less weight during the last 3 months of the experiment than at any time during the first 9 months (Table 32).

TABLE 32

Average increase in weight of each bread group over the four successive 3-month periods

Extraction rate of flour (per cent)	Average increase in weight (kg.) during:			
	1st 3 months	2nd 3 months	3rd 3 months	4th 3 months
Duisburg				
100	1·44	1·31	1·06	0·56
85	1·37	0·98	1·30	0·47
70	1·78	1·13	1·37	0·67
70 enriched to 100	1·68	1·19	1·19	0·57
70 enriched to 85	1·69	0·86	1·84	0·52
Vohwinkel				
100	1·53	1·15	1·24	1·00
70	1·85	0·85	0·64	1·00
70 enriched to 100	1·82	1·18	0·97	0·72

The group having unenriched white flour did not do worse than the others. Any failure to grow, therefore, was not due to lack of something which was supplied by one type of bread but not by another.

Twenty-one boys and 13 girls at Duisburg continued to eat the same type of diet for a further 6 months (see pp. 51, 52). The average height and weight of these 34 children at fortnightly intervals throughout 18 months are illustrated in Figs. 11 and 12.

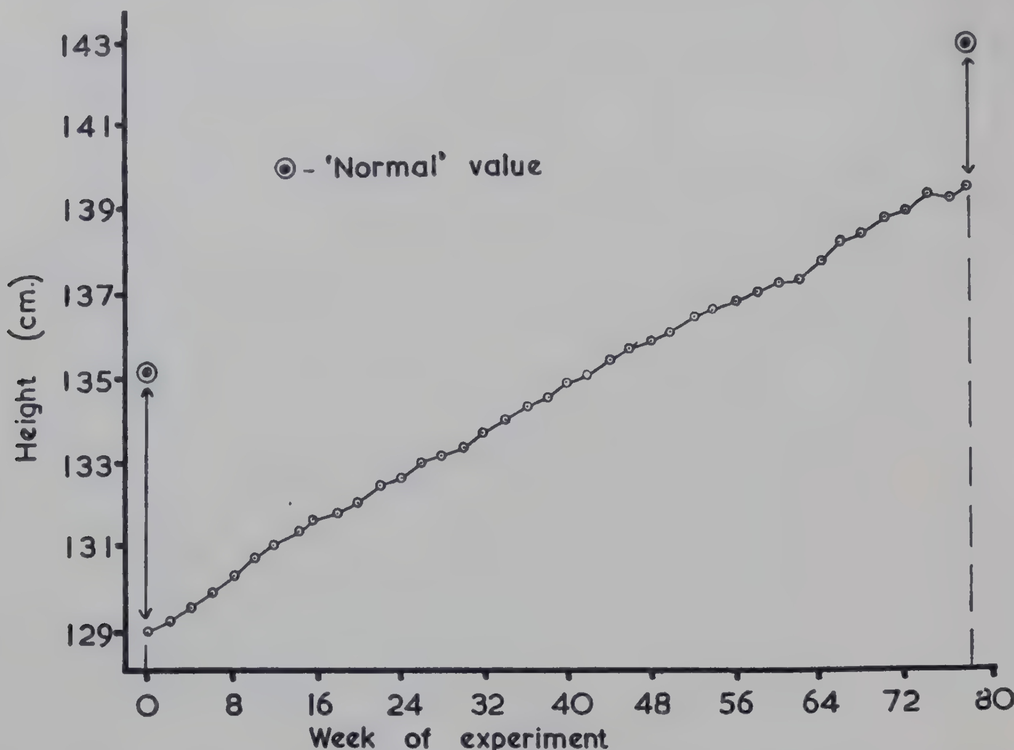

FIG. 11. Average height of 34 children at Duisburg who continued to eat the diets for a further 6 months.

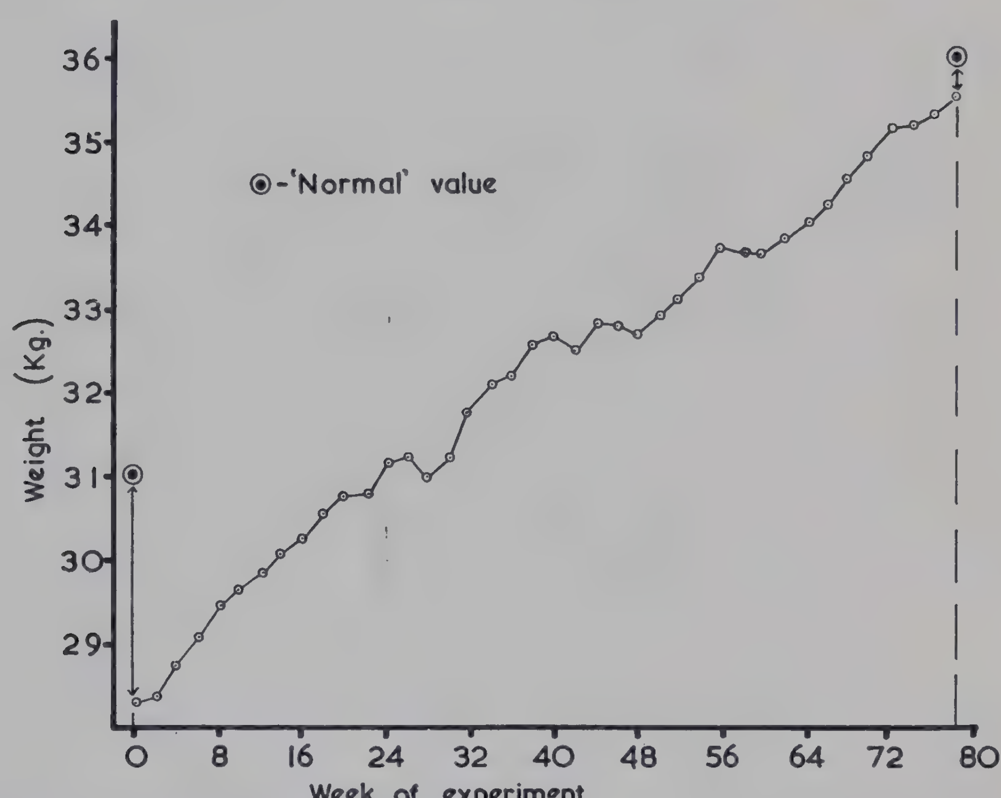

FIG. 12. Average weight of 34 children at Duisburg who continued to eat the diets for a further 6 months.

These children, whose average age at the beginning was 9 years 10 months, showed the same trends in growth rates during the first four quarters as the larger number of children already discussed (Table 33). During the last two quarters their rate of growth in weight increased again, however, so that during

TABLE 33

Calorie intakes and gains in height and weight of 34 children at Duisburg who took part in the experiment already described, and who continued to eat the same diet for a further 6 months

Measurement	Calorie intake and gain in height and weight during:					
	1st 3 months	2nd 3 months	3rd 3 months	4th 3 months	5th 3 months	6th 3 months
Average daily intake of Calories ..	2,157	2,175	2,174	2,106	2,321	2,159
Gain in height (cm.)	2·24	1·70	1·73	1·72	1·44	1·67
Gain in weight (kg.)	1·71	1·19	1·42	0·45	1·01	1·46
Gain in weight (g. per 1,000 Calories)	8·33	4·73	7·70	2·92	4·61	6·94

the last 3 of the 18 months they gained more weight than they had done at any time except during the first 3 months of the experiment, although by the end their weights were almost up to 'normal'. Had a dietary deficiency been developing at the end of the first year it is unlikely that the children would have gone on to increase their gain in weight again during the next 6 months.

SOMATIC MEASUREMENTS

The figures shown in Table 34 were arrived at in the following way. The average of each measurement was calculated for each of the 9 yearly age groups, 6–7, 7–8, 8–9, 9–10, 10–11, 11–12, 12–13, 13–14 and 14–15 years, both at the beginning and at the end of the investigation. These 9 average figures were then themselves averaged, and the final average at the beginning was compared with the final average at the end. For all measurements both for boys and girls at both

TABLE 34

Percentage increase in the somatic measurements of children aged 6 years 0 months to 15 years 0 months at the beginning of the experiments and those of children of the same age distribution at the end

Measurement	Percentage increase			
	Duisburg		Vohwinkel	
	Boys	Girls	Boys	Girls
Circumference of upper arm ..	3·7	5·9	3·5	3·6
Circumference of forearm ..	4·1	5·5	2·6	4·1
Circumference of thigh ..	2·3	4·0	1·8	2·4
Circumference of calf	3·4	4·7	2·8	4·3
Circumference outside deltoid	1·8	2·9	2·0	2·8
Circumference of chest ..	2·1	3·0	0·9	1·7
Circumference at iliac crests ..	2·2	3·9	1·9	1·5

homes the value at the end was greater than that at the beginning of the year. The kind of bread eaten did not make any detectable difference to the increase in the measurements, so in the presentation of the results all the children have been taken together. Table 34 shows the amount by which the measurements had increased, assuming the initial one to have been 100. It should be emphasized that this percentage increase does not represent the growth over the year; it represents the amount by which the measurements of children 6–15 years old at the end of the experiments exceeded those of children 6–15 years old at the beginning.

It will be seen that the measurements for girls showed a greater percentage increase than the corresponding measurements for boys, which is in accordance with their progress as regards height and weight (Table 24, p. 38). The measurements of the children at Duisburg tended to show a greater increase than those of the children at Vohwinkel. The results, taken as a whole, confirm the conclusion that during the year of experimental diets the children grew more rapidly than they had been growing in previous years. Figures not being available for 'normal' children, no comparisons with them can be made.

INTAKES, ABSORPTIONS AND EXCRETIONS

Tables 35 to 39 show the averaged results for the intakes, absorptions and excretions of nitrogen, calcium, phosphorus, magnesium, aneurin, riboflavin, and nicotinic acid and its derivatives, of the children eating the experimental diets. The numbers of children taking part in these experiments, the methods employed, the detailed findings and a discussion of them are given by Widdowson and Thrussell (p. 82) and by Holman (p. 92). The figures for all the children eating 70 per cent extraction flour, enriched or unenriched, have been averaged for nitrogen and minerals since there was no difference between these breads in respect of these elements.

The intakes of nitrogen from the different flours were not quite the same (see Tables 7 and 9), but the amounts excreted by the bowel were also different (Table 35), and the retentions were similar in all groups. It was part of the experimental plan that the intakes of nitrogen from bread, and consequently of total nitrogen, should be lower at Vohwinkel than at Duisburg (p. 17), but the children evidently obtained sufficient nitrogen from all the diets.

Table 36 shows the intakes of calcium, phosphorus and magnesium from bread and other foods and their routes of excretion. The 100 per cent extraction flour had been fortified with the most calcium, and the 70 per cent extraction flours with the least (see p. 13); this explains the difference between the intakes of the children eating them, and why the calcium-phosphorus ratio in the foods was approximately the same whatever the kind of bread. It will be seen that *at these levels of intake* the absorptions were almost exactly the same on each kind of bread and the balances satisfactorily positive. If the amount of calcium absorbed is expressed as a percentage of the intake, the absorption naturally appears smaller the higher the extraction rate of the flour.

In the case of phosphorus the higher intake on the higher extraction flours was due to phytic acid and its breakdown products, and not to added phosphorus. A little more phosphorus was absorbed from the higher extraction flours, but similar amounts were retained at each level of extraction. The calcium which had been added to the higher extraction flours probably prevented the absorption of some of the phosphorus in them. The ratio of the amount of

calcium to the amount of phosphorus retained ranged from 1·25 to 2·1. This suggests that satisfactory bone formation was going on whatever the extraction rate of the flour.

TABLE 35

Absorption and excretion of nitrogen on the experimental diets

traction rate of flour (per cent)	Intake (g. per day)			Excretion (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Urinary excretion as per cent of intake
	Bread and flour	Other foods	Total	Urine	Faeces	Total				
Duisburg										
100	11·1	3·7	14·8	9·5	3·1	12·6	+ 2·2	11·7	79·1	64·2
85	9·1	3·6	12·7	9·3	1·7	11·0	+ 1·7	10·9	85·8	73·2
) enriched and nriched together	9·5	3·8	13·3	9·5	1·6	11·1	+ 2·2	11·7	88·0	71·4
Vohwinkel										
100	5·9	5·3	11·2	6·8	2·5	9·3	+ 1·9	8·7	77·7	60·7
) enriched and nriched together	5·5	5·3	10·8	6·7	2·0	8·7	+ 2·1	8·8	81·5	62·0

TABLE 36

Absorption and excretion of calcium, phosphorus and magnesium on the experimental diets at Duisburg

traction rate of flour (per cent)	Intake (g. per day)			Excretion (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Urinary excretion as per cent of intake
	Bread and flour	Other foods	Total	Urine	Faeces	Total				
Calcium										
100	1·90	0·62	2·52	0·22	2·07	2·29	+ 0·23	0·45	17·9	8·7
85	0·85	0·61	1·46	0·23	1·02	1·25	+ 0·21	0·44	30·1	15·8
'0 enriched and enriched together	0·59	0·64	1·23	0·24	0·68	0·92	+ 0·31	0·55	44·7	19·5
Phosphorus										
100	1·68	0·46	2·14	0·68	1·32	2·00	+ 0·14	0·82	38·3	31·8
85	0·91	0·46	1·37	0·62	0·59	1·21	+ 0·16	0·78	56·9	45·3
'0 enriched and enriched together	0·60	0·48	1·08	0·56	0·37	0·93	+ 0·15	0·71	65·7	51·9
Magnesium										
100	0·68	0·13	0·81	0·15	0·63	0·78	+ 0·03	0·18	21·9	18·7
85	0·32	0·13	0·45	0·13	0·28	0·41	+ 0·04	0·17	37·6	30·2
'0 enriched and enriched together	0·17	0·14	0·31	0·12	0·16	0·28	+ 0·03	0·15	46·9	37·2

The magnesium in breads of high extraction is largely present as the phytate and hence the variations in the amounts of magnesium ingested vary with those of phosphorus. The absorptions, paths of excretion and retentions of magnesium

TABLE 37

Intake and excretion of aneurin on the experimental diets

Extraction rate of flour (per cent)	Intake (mg. per day)			Total intake (mg. per 1,000 Calories)		Excretion (mg. per day)			Urinary excretion as per cent of intake
	Bread and flour	Other foods	Total	Total Calories	Non-fat Calories	Urine	Faeces	Total	
Duisburg									
100	1·64	0·60	2·24	0·92	1·00	0·61	0·65	1·26	27
85	1·27	0·59	1·86	0·86	0·95	0·39	0·72	1·11	21
70	0·62	0·65	1·27	0·55	0·59	0·17	0·46	0·63	13
70 enriched to 100	1·40	0·62	2·02	0·84	0·91	0·67	0·69	1·36	33
70 enriched to 85	1·23	0·60	1·83	0·82	0·90	0·37	0·72	1·09	20
Vohwinkel									
100	0·84	0·75	1·59	0·57	0·89	0·51	0·53	1·04	32
70	0·32	0·75	1·07	0·41	0·62	0·19	0·28	0·47	18
70 enriched to 100	0·82	0·76	1·58	0·55	0·84	0·40	0·20	0·60	25

TABLE 38

Intake and excretion of riboflavin on the experimental diets

Extraction rate of flour (per cent)	Intake (mg. per day)			Total intake (mg. per 1,000 Calories)	Excretion (mg. per day)			Urinary excretion as per cent of intake
	Bread and flour	Other foods	Total		Urine	Faeces	Total	
Duisburg								
100	0·93	0·39	1·32	0·54	0·23	2·58	2·81	17
85	0·52	0·38	0·90	0·41	0·25	1·42	1·67	28
70	0·23	0·37	0·60	0·26	0·11	1·49	1·60	18
70 enriched to 100	0·55	0·41	0·95	0·40	0·28	1·47	1·75	30
70 enriched to 85	0·46	0·39	0·85	0·39	0·28	1·23	1·51	33
Vohwinkel								
100	0·41	0·93	1·34	0·48	0·07	0·59	0·66	5
70	0·12	0·90	1·02	0·39	0·07	0·59	0·66	7
70 enriched to 100	0·25	0·84	1·09	0·38	0·17	0·68	0·85	16

also resembled those of phosphorus in that the amounts absorbed and retained were very much the same whatever the kind of bread.

The intakes of aneurin and the urinary excretions varied with the extraction rate or the enrichment of the flour (Table 37); foods other than bread and flour provided 0·6 to 0·75 mg. per day, which is a relatively large amount (Holman, p. 92). The total intakes at both homes, even on the diets containing unenriched white flour, were equal to or greater than the highest value for the requirement which has been proposed. Although the intakes from bread and flour were deliberately made lower at Vohwinkel than at Duisburg, the intake from other foods was higher and consequently the total intakes were not as different as had been expected.

The intakes of riboflavin (Table 38) were barely sufficient if some of the estimates for the requirement (0·5 mg. per 1,000 Calories) are correct. The excretions in the urine, however, do not suggest that there was any deficiency (see Holman, p. 92).

It is possible that some of the children at Vohwinkel who were eating the unenriched white flour might have shown some signs of a deficiency had their other foods provided no more riboflavin than the Duisburg 'basal' diet. The German rations at Vohwinkel, however, provided so much riboflavin that the total intake was higher than at Duisburg in spite of the fact that the children were deriving one third of their calories from fat and sugar.

Children eating bread made from flour of 100 per cent extraction tended to excrete smaller amounts of riboflavin in the urine than children eating the corresponding enriched bread.

TABLE 39

Intake and excretion of nicotinic acid derivatives on the experimental diets

xtraction rate of flour (per cent)	Intake (mg. per day)			Total intake (mg. per 1,000 Calories)	Excretion (mg. per day)				Total* urinary excretion as per cent of intake
	Bread and flour	Other foods	Total		Urinary acid-hydrolysable nicotinic acid	Urinary N-methyl-nicotinamide	Urinary pyridone	Nicotinic acid in faeces	
Duisburg									
100	27·9	8·0	35·9	14·7	0·9	7·0	—	17·0	20
85	13·3	7·8	21·1	9·8	1·0	4·8	—	6·1	25
70	8·1	7·2	15·3	6·8	0·8	5·2	—	2·7	36
0 enriched to 100	25·4	8·4	33·8	14·0	1·2	9·8	—	7·5	30
'0 enriched to 85	15·4	8·0	23·4	10·6	0·9	6·6	—	2·3	29
Vohwinkel									
100	14·0	12·7	26·7	9·6	1·2	6·1	6·3	6·1	44
70	5·8	10·8	16·6	6·4	1·2	4·1	5·0	8·5	54
70 enriched to 100	14·1	12·6	26·7	9·3	1·3	6·5	12·4	8·6	64

* Expressed as nicotinic acid. The Duisburg results do not include the pyridone.

The faeces of the children at Duisburg always contained more riboflavin than the food. This must have been the result of biosynthesis.

The intakes of nicotinic acid (Table 39) exceeded the U.S. National Research Council allowance of 4·7–5·0 mg. per 1,000 Calories in all groups at both homes. Judging by the levels of excretion of N-methylnicotinamide and of pyridone it appears probable that the intakes in all the groups were adequate. There is a suggestion from the quantities of pyridone excreted by the children at Vohwinkel that more nicotinic acid was absorbed from the enriched flour than from the flour of 100 per cent extraction.

Discussion

These experiments were undertaken to study the effect of different extraction rates and of enrichment with B-vitamins and iron on the nutritional value of wheaten bread for children. The study was confined to the proteins in the wheat and to the vitamins of the B complex. No difference could be detected between the nutritional values of the breads and, since the children grew at a very satisfactory rate and no signs of any deficiencies appeared, it may be concluded that all the diets supplied the children with the nutrients known and unknown which they required for the period over which the experiments were conducted.

The children were about 5 per cent below American standards for height and 8 per cent below for weight when the experiments began and it was thought at the time that this made them good subjects for the investigation. It has been suggested, however, that the very fact that they were undernourished might have been the reason why the breads gave equally satisfactory results; for it is known that undernourished animals may make a good initial response to a supplement which ultimately fails to provide them with all they require. If this is so, it can only affect the interpretation of the results (see Irwin, p. 71).

It cannot be emphasized too strongly that the results themselves apply only to the particular conditions under which the investigation was carried out. The diets contained more vegetables, but much less milk and meat, and in one experiment less fat and sugar, than diets in use at the present time in Britain, so that conclusions are in any case not directly applicable to this country.

Dutch investigators (Reith, Gorter and van Eekelen, 1949; van Eekelen, 1949) have studied the effect of the extraction rate upon the nutritional value of diets commonly in use in Holland. They have approached the problem by calculation rather than by experiment, and have shown that, whatever the extraction rate, the quantity of the B-vitamins and of the essential amino-acids in the total diet exceeds the requirements of these materials as assessed by the National Research Council Food and Nutrition Board (1948). The validity of their conclusions clearly depends upon a correct assessment of requirements. Engel, Gorter and van Eekelen (1951) have checked these conclusions by experiments on rats which were given "a normal mixed Dutch diet for human consumption, in which bread furnished 30–35 per cent of the total calories". The rats having white bread (75 per cent extraction) grew just as well as those having whole wheat bread.

PART II: AN EXPERIMENT WITH A MILK SUPPLEMENT

THE rate at which a child grows depends to a great extent upon how good and plentiful its food is, and, since the experiment of Corry Mann (1926), many people have improved the growth rates of children by adding a milk supplement to their customary diets. As a result the idea has grown up that milk is an essential for good nutrition, and that only a diet which cannot be made to promote better growth by supplementing it with milk can be considered fully adequate.

The excellent growth displayed by the children at Duisburg on diets to which wheat contributed 75 per cent of the calories, and which contained very little milk or animal protein, raised the question as to whether this growth could be improved still further by milk. Current ideas about the food requirements of growing children strongly suggested that it could; yet the growth rates at Duisburg were already so good that any further improvement seemed, on the face of it, likely to be difficult to achieve. The stage seemed to be set for an interesting and important experiment. This was accordingly undertaken.

The Organization of the Experiment

THE SUBJECTS

All the children over 5 years of age who were living in the orphanage at Duisburg in June, 1948, were included in this investigation. Some of them had taken part for the full 12 months in the experiment on extraction rates already described; others had been admitted to the home at some time during the first 6 months of that investigation. The latter had partaken of the same experimental diets as the other children, and they had been examined, weighed and measured along with them, but they were not included when the results were being presented because they had been there for too short a time. A third group was composed of those who had entered the orphanage during the second 6 months of the investigation. They had not eaten the experimental breads, but had been able to eat unlimited quantities of German bread because this had been obtained as part of the official rations issued to all the children in the home, whether they had been eating the experimental diets or not.

The children were divided into two groups, as alike as possible as regards age, sex, height, weight and clinical assessment, and also as regards their previous history, i.e. whether they had previously taken part in the comparison of the extraction rates or not, and if so, for how long. The two groups included equal numbers of children from each of the five 'bread' groups.

The investigation lasted for 6 months, from June to December, 1948. Table 40 shows the number of boys and girls in the three categories who started and who completed the full 6 months of the investigation now being described. The findings for all the children who remained for the whole 6 months have been put together in the presentation of the results.

All the children were weighed and measured every fortnight during the investigation as described on p. 8. They were examined clinically before the experiment started (see p. 7) and after 3 and 6 months, but blood was not taken, nor were any radiological examinations made.

The average starting ages of the children in the two groups who completed the experiment, and their initial heights and weights expressed as a percentage

TABLE 40

Numbers of children who started and who completed the 6 months of the experiment

Previous history	Group	Numbers starting		Numbers completing 6 months	
		Boys	Girls	Boys	Girls
Children who had completed 12 months of the experiment on extraction rates	Milk	24	22	21	22
	Control	24	20	21	13
Children who had taken part in the experiment on extraction rates for 6–12 months . .	Milk	4	10	3	4
	Control	4	10	3	10
Children who had been in the home for 2–6 months but had not taken part in the experiment on extraction rates . .	Milk	15	12	11	6
	Control	15	13	11	10
Total	Milk	43	44	35	32
	Control	43	43	35	33

TABLE 41

Average ages of the children at the beginning of the experiment, and their heights and weights expressed as a percentage of their 'normal' heights and weights calculated from the tables of O'Brien, Girshick and Hunt (1941)

Group		Average age at beginning (years and months)	Average height as per cent of 'normal' at beginning	Average weight as per cent of 'normal' at beginning
Milk:	Boys . .	10–1	96·4	97·1
	Girls . .	10–3	96·1	95·3
Control:	Boys . .	10–1	97·0	95·7
	Girls . .	10–0	95·8	96·2

of 'normal' (p. 5), are given in Table 41. If the values for heights and weights are compared with those in Tables 2 and 6 (pp. 5, 12) it will be seen that, as would be expected, the children were now nearer to 'normal' dimensions.

THE DIETS

Since it had already been shown that children did equally well whichever bread they ate, it was decided to use only one kind of flour for making the bread for this experiment. Flour of 85 per cent extraction was chosen because this was

in general use in England at the time and was most readily available. All the children in both groups had their German rations except for the cereals, and they all had unlimited amounts of bread made from flour of 85 per cent extraction fortified with calcium carbonate as described on p. 13. As before, sufficient jam was supplied to spread sparingly on the bread. One group, the milk group, received 500 ml. of whole milk during the day, reconstituted from full-cream milk powder. This was reckoned to provide 330 Calories and was additional to the small quantities of milk which were included in the German ration. It was given in two portions, 250 ml. in the middle of the morning when the children came in from school (see p. 15), and 250 ml. when they came back from school in the afternoon.

The children in the control group had only the milk provided by their German rations, which amounted to 60–120 ml. a day. When they came back from school in the middle of the morning they were given plain biscuits, made from the experimental flour with sugar and margarine, together with a drink of orange juice, made up from concentrated orange juice and water. They had a similar 'snack' when they came in in the afternoon, and the total energy value of their supplement was 330 Calories per day, the same as that provided by the milk. Sufficient calcium carbonate was added to the orange juice to make the calcium intakes of the children in the control group equal to those of the children in the milk group, but this was probably of no experimental importance because the children's bread was fortified with calcium to the extent of 0·15 g. per 100 g. flour (see p. 13). It will be noted that the supplements were given apart from the main meals, and that the latter were identical for both groups of children. Quantitative dietary records were kept as described on p. 15.

Throughout the experiment all the children received vitamin tablets providing daily 2,000 I.U. of vitamin A, 1,000 I.U. of vitamin D and 25 mg. of ascorbic acid.

Table 42 shows the average composition of the diets eaten by the two groups of children over the whole 6 months of the experiment. The larger amount of

TABLE 42

Composition of the diets

Constituent of diet	Milk group	Control group
Total protein (g. per day)	72·6	61·4
Animal protein (g. per day) ..	26·5	8·8
Fat (g. per day)	45·6	40·8
Carbohydrate (g. per day).. ..	346·0	371·0
Percentage of total calories from:		
protein	13·9	11·7
fat	19·8	17·7
carbohydrate	66·3	70·6
Calcium (g. per day)	1·83	1·3 + 0·53 added to orange juice
Phosphorus (g. per day)	1·60	1·19
Iron (mg. per day)	12·9	14·0

fat in the control diet as compared with the figure given in Table 9 (p. 17) is accounted for by the margarine used in making the biscuits. In other respects

the diet was very similar to that eaten by the children during the study of extraction rates. The supplement of milk trebled the intake of animal protein.

The average calorie intakes over the whole period are shown in Table 43. Whether they are expressed as total calories or as calories per kg. of body weight the figures for the two groups are remarkably similar, and it is clear that the milk supplement did not specifically increase the children's appetites. For the first few weeks of the experiment, however, *all* the children ate more food, whether they were getting the extra milk or not. Table 44 shows the average

TABLE 43

Average daily calorie intakes over the 6 months of the experiment

Group	Total Calories		Calories per kg. body weight	
	Boys	Girls	Boys	Girls
Milk ..	2,240	2,017	71·0	63·0
Control ..	2,258	2,054	72·0	64·6

TABLE 44

Average daily calorie intakes of boys and girls at the end of the experiment on extraction rates and at the beginning of the milk experiment

Period	Average daily Calories:			
	Milk group 23 boys	Control group 24 boys	Milk group 26 girls	Control group 23 girls
Last 4 weeks of experiment on extraction rates	2,154	2,054	1,987	2,040
First 4 weeks of milk experiment	2,457	2,423	2,264	2,328
Second 4 weeks of milk experiment	2,262	2,215	2,053	2,106
Third 4 weeks of milk experiment	2,282	2,266	2,026	2,107

daily calorie intakes of those children who partook of the experimental diets during the last 4 weeks of the experiment on extraction rates, and who went on for the first 12 weeks of the milk experiment. During the first 4 weeks of this experiment they seemed to take their supplements of milk or of biscuits in addition to their accustomed amounts of bread. During the second 4 weeks they ceased to do so, at any rate to the same extent, and they maintained their calorie intakes at the lower level until the end of the experiment. It is not known why the children ate more food. It may have been the novelty of the supplements,

which were not given at the main meals so that the children could still indulge in these to the limits of their capacity. It may have been that children of the same sex and of similar ages were once again feeding together as they had been accustomed to do before the study of extraction rates began. This led to a certain amount of rivalry, particularly among the older boys, as to who could eat the greatest number of slices of bread.

Results

CLINICAL EXAMINATIONS

Clinical Grading

Since most of the children who took part in this experiment had already participated in the work on extraction rates, they had already improved greatly in physique, and a high proportion of the total number was graded A at the outset. By the end of the 6 months of the milk experiment, 6 of the children who had been having supplementary milk were given a higher grading than at the beginning. Of the control group 7 were upgraded and 2 deteriorated slightly. These 2 happened to be unwell at the time of the final examination.

Skin Changes

There was no further decrease in the incidence of skin sepsis in either group during the 6 months of the milk experiment. Hyperkeratosis pilaris, however, continued to diminish in spite of the fact that the initial examination was made in June and the final one in December (McCance and Barrett, 1951). Eleven children receiving supplementary milk and 18 children in the control group had hyperkeratosis at the beginning. Only 6 having milk and 7 of the control group had any at the end.

Muscular Tone and Development, and Subcutaneous Fat

Muscular tone and development and the amount of subcutaneous fat continued to improve in both groups, and there was as much improvement in the control group as in the group receiving additional milk.

Thyroid Enlargement

All the children in both groups continued to receive supplementary iodine (see p. 26) throughout the 6 months of the experiment. When the experiment began 18 children had conspicuously enlarged thyroid glands and 54 slightly enlarged glands. At the end 16 thyroids were conspicuously enlarged and 63 slightly enlarged. There was therefore very little change in the size of the thyroid glands. There was no evidence that the additional milk made any difference to the size of the glands.

HEIGHTS AND WEIGHTS

Fig. 13 shows the average gains in height and weight of the children in the two groups over the 6 months of the investigation. The curves for the two groups could hardly be more alike, and it is evident that the children having the supplement of milk did not grow any faster than those not having it. The rapid gains in weight over the first 4 weeks of the experiment may be explained by the temporary increase in calorie intake during that time (see p. 54), but if so the levelling off during the second 4 weeks is difficult to explain, for the

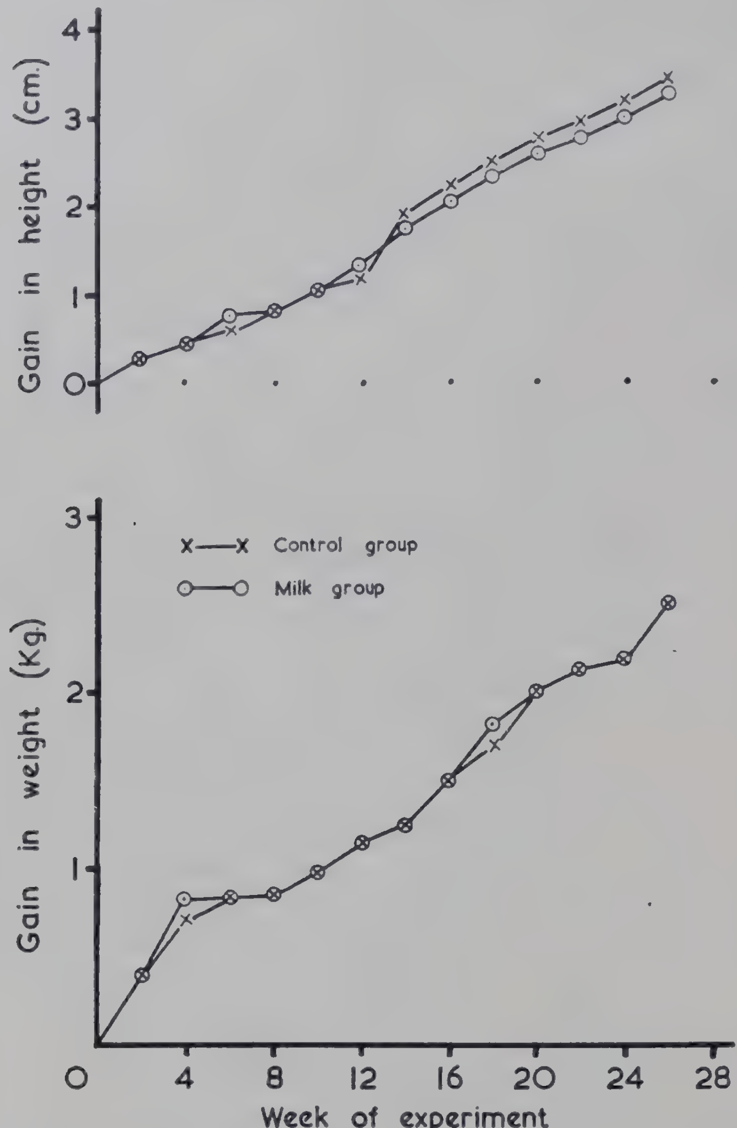

FIG. 13. Average gains in height and weight of the children in the milk and control groups over the 6 months of the investigation.

TABLE 45

Growth in height and weight expressed as a ratio of the actual increase to the 'normal' increase

Group		Ratio of actual increase in growth to 'normal' increase	
		Height	Weight
Milk:	Boys	1·18	1·38
	Girls	1·40	1·57
	All children ..	1·28	1·47
Control:	Boys	1·15	1·32
	Girls	1·45	1·62
	All children ..	1·29	1·47

growth rates accelerated again after the 8th week without any change in calorie intake. Both groups gained more height and weight than 'normal', for the 'normal' gain in height over the 6 months for a group of children of similar age and sex to those taking part in the experiment is 2·6 cm. and in weight 1·65 kg.

The ratio of the actual increase in height and weight to the 'normal' increase is given in Table 45. This table brings out once again the close similarity between the results for the two groups. As in the study of extraction rates (Table 24, p. 38) the girls gained more than the boys.

When the gains are expressed as a percentage of the initial heights and weights (Table 46) the same similarity is seen.

TABLE 46

Growth in height and weight during the 6 months of the experiment

Group		Average height at beginning (cm.)	Gain as per cent of starting height
Milk:	Boys	131·4	2·4
	Girls	131·7	2·6
	All children ..	131·5	2·5
Control:	Boys	132·3	2·4
	Girls	130·0	2·6
	All children ..	131·1	2·5
		Average weight at beginning (kg.)	Gain as per cent of starting weight
Milk:	Boys	30·4	7·9
	Girls	30·7	8·5
	All children ..	30·5	8·2
Control:	Boys	30·1	8·0
	Girls	30·5	8·2
	All children ..	30·3	8·1

TABLE 47

Average gain in weight per 1,000 *Calories and per g. of protein in the diet*

Group		Gain per 1,000 Calories (g.)	Gain per g. protein (g.)
Milk:	Boys ..	5·82	0·17
	Girls ..	7·05	0·21
Control:	Boys ..	5·80	0·19
	Girls ..	6·67	0·23

Table 47 shows the gains in weight calculated per 1,000 Calories and per g. of protein in the diet. Since the calorie intakes and the total gains were so alike for the two groups, the gains per 1,000 Calories were also similar. The gains per g. of protein, however, were a little less in the milk group because the protein intake was higher.

Discussion

Why were the present results so different from those of previous investigators? In other words, why have previous investigators almost invariably increased the growth rate of children by supplementing their diets with milk, while we have not? The difference must lie either in the quality or quantity of the basal diet, or in the composition of the dietary supplement with which the milk supplement was compared. Milk (500 ml.) provided 330 Calories, 17 g. animal protein, 18 g. fat and 0·6 g. calcium, as well as other important substances such as vitamin B_{12}. If the basal diet had been short of one or more of these constituents, supplementing it with milk or with any other food which made good the deficiency would have been likely to increase the growth rate.

The work of previous investigators may be considered according to the way in which their experiments were designed. Clark (1929), Turbott and Rolland (1932), Scharff and Sinnadorai (1937), Loewenthal (1938), Roberts, Blair, Lenning and Scott (1938) and the Milk Nutrition Committee (1939) gave a milk supplement to one group of children and gave no supplement, or only a placebo, to a second group. The better growth of the children having milk was interpreted as showing the special value of milk, but many other foods might have given just as good a response, and all that can be said with certainty is that the various basal diets must have been unappetizing or nutritionally unsatisfactory. Orr (1928) and Leighton and Clark (1929) controlled their supplement of whole milk by giving an equal volume of skimmed milk to one group, and biscuits equal in calorie value to the skimmed milk to another; a fourth group received no supplement. The two groups receiving milk grew equally well and better than the other two groups, between which there was also no difference. The children were living in their own homes and the investigators did not know the make-up of the basal diets, but the results suggest that these were not deficient in calories. The milk may have made good a deficiency of animal protein or of calcium. Aykroyd and Krishnan (1939) gave supplements of 30 g. of skimmed milk powder or 1 g. of calcium lactate a day to Indian schoolboys who were living on a diet consisting chiefly of rice. The boys in the milk group grew a little better than those in the calcium lactate group, and both groups grew better than a control group of boys who received a placebo. It would appear that in this investigation calcium was one of the effective ingredients in milk. There may have been others but it is impossible to tell, since the milk supplement itself contained nearly three times as much calcium as the supplement of calcium lactate.

The pioneer experiment on this subject was that of Corry Mann (1926). He compared the effects of equi-calorific amounts of milk, butter, sugar and margarine, and he controlled the animal protein in the milk by giving to another group a supplement of casein; a sixth group was given watercress as a source of vitamin A and a seventh group received no supplement. His results are a little difficult to interpret when examined critically, but it would appear that the group receiving milk grew better than any of the others, and that this was not due to the protein in the milk. It may have been due partly to its calorie value,

but it seems likely that calcium was the important ingredient, for it is clear from the information given by Corry Mann that the basal diets provided very little of this element, and that milk was the only one of his dietary supplements which contained much calcium. The basal diet at Duisburg supplied the children with unlimited calories and all the calcium and vitamin D they can have required. The present investigation is, in fact, the only one in which a milk supplement has been added to a diet known to contain plenty of calcium and vitamin D.

The basal diet at Duisburg seemed to be critically short of animal protein, and it was expected that if the milk were going to have an effect it would be because it corrected this. No attempt was made to balance this contribution in the milk, for the biscuits given to the control children merely added a little more cereal protein to a diet already providing this ingredient in generous quantities. It must be concluded that the mixture of amino-acids which the children obtained from their wheat flour and vegetables was not deficient in any respect, although it is possible that the small amounts of animal foods in the basal diet were of vital importance, particularly as a source of vitamin B_{12}. The fact that milk did not promote better growth shows that the experimental diets were wholly satisfactory for these children.

PART III: ANIMAL EXPERIMENTS

When the experiments on the children failed to show that flour of any one extraction rate promoted better growth than flour of any other, it seemed very desirable to make similar experiments with animals on which wholemeal bread had previously been demonstrated to be superior in this respect (Chick, 1940, 1942; Childs, Macrae, Braude and Kon, 1948).

Experimental

Rats

Black and white hooded rats from the inbred colony of the Department of Experimental Medicine, Cambridge, were used for these experiments. Most of the work was done on males, but one study of females was made. The rats were weighed and grouped at weaning, and in all the experiments each group contained 6 rats. Each animal was weighed weekly. The experimental diets consisted of mixtures made up to resemble the diets eaten at the two orphanages (see Table 8, p. 16). Breads made from 100 per cent and 70 per cent extraction flours fortified with calcium carbonate were used. The four kinds of diets, which were prepared from fresh foods each day and were provided *ad lib.*, were minced so that the rats were forced to eat the mixture and could not pick out the bits they liked best. A record was kept of the amounts of the mixture eaten by each group of rats each day. For every experiment a control group of 6 rats was fed on a stock diet, consisting of a mixture of bread, yeast, whole wheat, oatflakes, bran, fishmeal, wheat germ and whole milk, with liver once a week, and greens every alternate day. The Duisburg diets contained 12–13 per cent of protein, the Vohwinkel diets 9–10 per cent, and the stock diet 19 per cent.

In some of the experiments the animals were given the experimental diets from weaning, in others they were first given the stock diet in limited or unlimited amounts for about 5 weeks and then were given the experimental diets. All the rats eating the experimental diets received supplements of vitamins A and D.

At the end of one of the experiments, which included females as well as males, the reproductive capacity of both sexes was tested. The rats were mated with other rats which had lived on the stock diet throughout their lives. The experimental males were mated by placing 2 of each group in cages of 6 non-experimental females and leaving them there for 5 days. Then these 2 males were removed and another 2 of the same group were put in for a further 5 days, and finally the last 2 for yet another 5 days. The experimental females were mated by placing 2 non-experimental males in each cage of 6 females and leaving them there for 15 days. A record was kept of the number of rats in each group which became pregnant, the numbers born alive and dead in each litter and the numbers successfully reared.

At the end of another experiment the rats were killed. The bodies were opened and spread out on trays, and they were dried in an oven at 100° C. to constant weight. Assuming the fat-free portion of the body to contain 73 per cent of water (McCance and Widdowson, 1951 b; Spray and Widdowson, 1951) and the fatty portion to contain none, the percentage of fat in the rats' bodies was then calculated from the percentage of water.

Pigs

Animals bred from Large White or first cross Wessex/Large White sows mated to a Wessex boar were used. The pigs were 9 – 11 weeks old when the

experiments started, and the males had already been castrated. Two experiments were made, and one litter of pigs was used for each experiment. Each kind of flour was fed to 2 animals. The pairs were chosen so that the combined starting weights of the 2 pigs in each pair were as nearly alike as possible. The pigs were weighed every fortnight. Again, mixtures were made up to simulate the diets eaten by the German children in the two orphanages (Table 8, p. 16) and these mixtures were fed to the pigs. As in the case of the rats, flours of 100 per cent and 70 per cent extraction containing added calcium carbonate were used and, since it was found that the pigs ate flour just as readily as bread, the flour itself was incorporated into their ration. The pigs also received supplements of vitamins A and D. A record was kept of the amount of food eaten by each pair of pigs each day.

The pigs were killed at the end of the experiments, and measurements and analyses of body fat were made according to the methods described by Walley (1952).

Results

THE EXPERIMENTS ON RATS

The Effect of giving the Experimental Diets to Rats from Weaning

Male and female rats (6 in each group) were allowed to eat unlimited amounts of the four experimental diets from weaning. A fifth group from each sex was fed on the stock diet. Figs. 14 and 15 show the average growth rates in the five

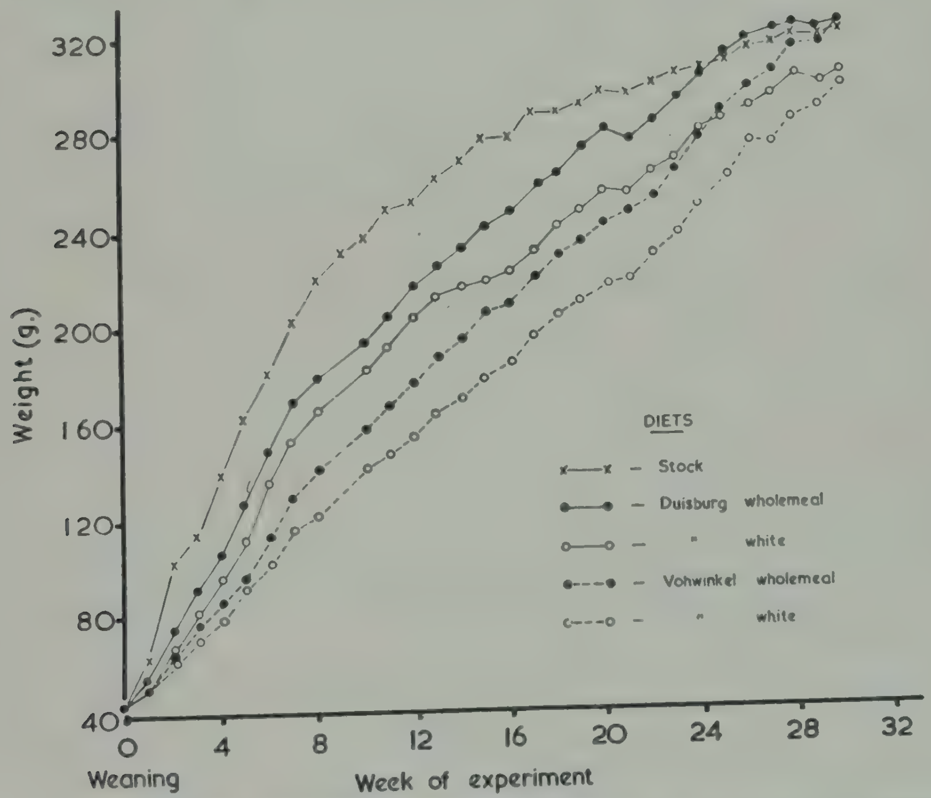

FIG. 14. Average growth rates of male rats given the experimental diets from weaning.

groups. The rats having wholemeal bread grew better than those having the corresponding white bread diet. Both brown and white bread groups on the

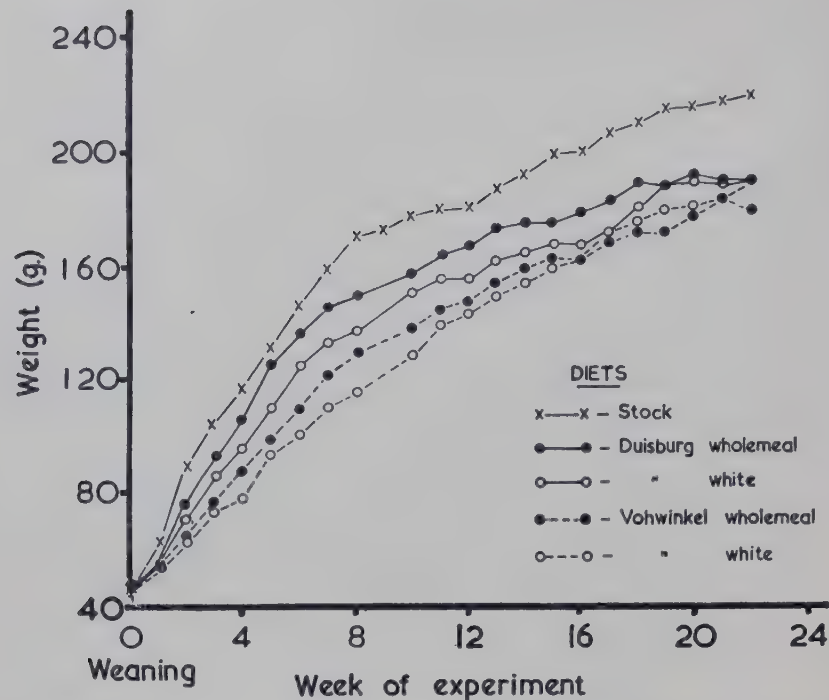

FIG. 15. Average growth rates of female rats given the experimental diets from weaning.

Duisburg diet gained more weight than either group having the Vohwinkel diet, although the latter ate 10 per cent more food in terms of calories. No groups grew as well as animals receiving the stock diet, which contained more protein.

After the experiment had been running for 22 weeks, five groups of male and female rats were mated with animals which had been reared on the stock diet. The reproductive capacity of the experimental male rats was normal, since all the non-experimental females with which they were mated became pregnant and produced healthy litters. The results for the experimental female rats are summarized in Table 48. At least 5 of the 6 rats in every group gave birth to

TABLE 48

Reproductive capacity of female rats which had lived on the experimental diets from weaning

(Each group consisted of 6 rats)

Diet and extraction rate of flour (per cent)	Number becoming pregnant	Total number born		Average weight at birth (g.)	Number of litters reared	Total number of rats reared	Average weight at weaning (g.)
		Alive	Dead				
Duisburg							
100	5	48	2	5·5	1	7	23
70	5	37	5	5·2	3	24	23
Vohwinkel							
100	5	16	5	6·3	2	9	23
70	6	42	4	5·3	2	14	20

litters, most of which were born alive, and were of normal weight. The mothers in none of the experimental groups, however, were able to rear their litters satisfactorily.

The Effect of starting the Experimental Diets at Eight Weeks of Age

For 5 weeks after weaning 30 male rats were given unlimited amounts of the stock diet, and were then given the experimental diets. The animals grew at the same rate on each kind of bread (Fig. 16). The similarity of the growth rates was probably due to the fact that the experiment was not started until the rats

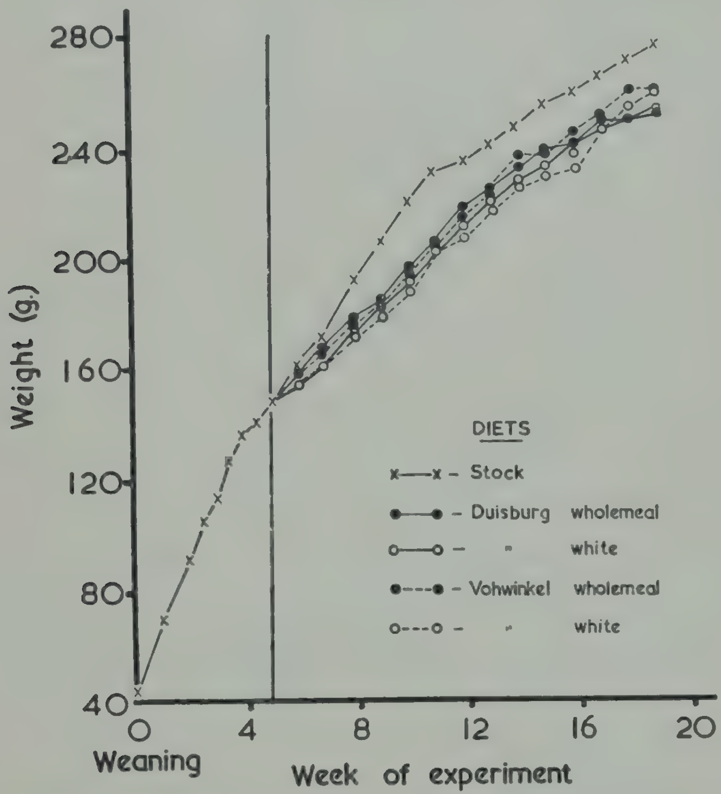

FIG. 16. Average growth rate of male rats given the experimental diets from 8 weeks of age.

were 8 weeks old, by which time they must have passed the critical period of their lives when some constituent of wholemeal flour was of vital importance for growth.

The rats having the Vohwinkel diets ate more in terms of calories than those having the Duisburg diets, and on both diets the animals having brown bread ate slightly more than those having the white. The gain in weight per 1,000 Calories, or per g. of protein, was slightly greater for those having the white bread.

The Effect of giving the Experimental Diets to Undernourished Male Rats Eight Weeks old

The object of this experiment was to reproduce more exactly the state of affairs at the orphanages. For 2 weeks after weaning, 30 male rats were given unlimited amounts of stock diet, and then their food was restricted so that their average weight at 8 weeks of age was only 107 g. The litter mates of these animals, which were used for the experiment shown in Fig. 16, weighed 148 g.

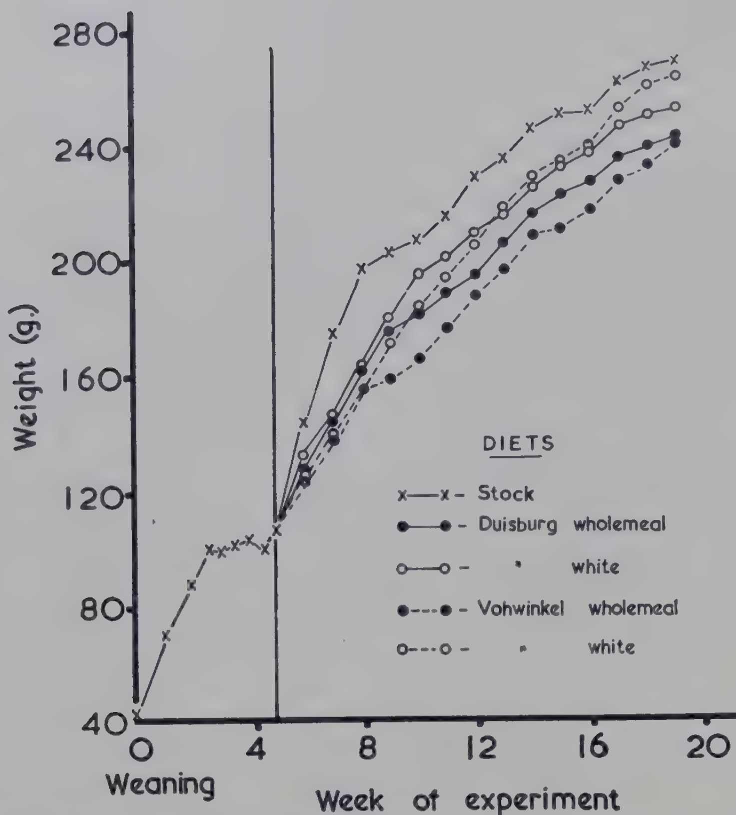

FIG. 17. Average growth rate of undernourished male rats given the experimental diets from 8 weeks of age.

at this time. The gains in weight of the rats on the stock diet and on the four experimental diets are shown in Fig. 17. All the groups grew very rapidly and equally well at the beginning of the experimental period. By the end, the two groups of animals having white bread were slightly heavier than the two groups of animals having brown.

A Comparison of the Amounts of Fat in the Bodies of Rats reared from Weaning on the Experimental Diets

The left half of Table 49 shows the composition of young rats whose growth rate had never been interrupted (see Fig. 14). The type of bread made no

TABLE 49

Composition of the bodies of male rats reared on the experimental diets

Diet and extraction rate of flour per cent	Animals aged 15 weeks		Animals aged 30 weeks	
	Non-fat tissue (g.)	Fat (g.)	Non-fat tissue (g.)	Fat (g.)
Duisburg				
100	233	46	—	—
70	222	40	224	49
Vohwinkel				
100	189	30	—	—
70	185	33	226	93

difference to the composition of the body. The 'Vohwinkel' animals were lighter than the 'Duisburg' animals. They contained less flesh and less fat but the ratio of fat to lean was very similar. The right half of the table shows the composition of mature animals whose growth had been checked by a period of undernutrition (see Fig. 17). The 'Vohwinkel' animals were heavier and this was entirely due to fat.

THE EXPERIMENTS ON PIGS

Growth

Preliminary tests showed that when pigs were allowed to feed freely on the experimental diets they tended to eat far more of the Vohwinkel diet than of the Duisburg one. In this experiment the Duisburg diets were provided *ad lib.*, and the Vohwinkel diets were not allowed to exceed them by more than 10 per cent in terms of calories. This reproduced the freely chosen intakes of the children at the two orphanages. Each curve in Fig. 18 represents the average weight of

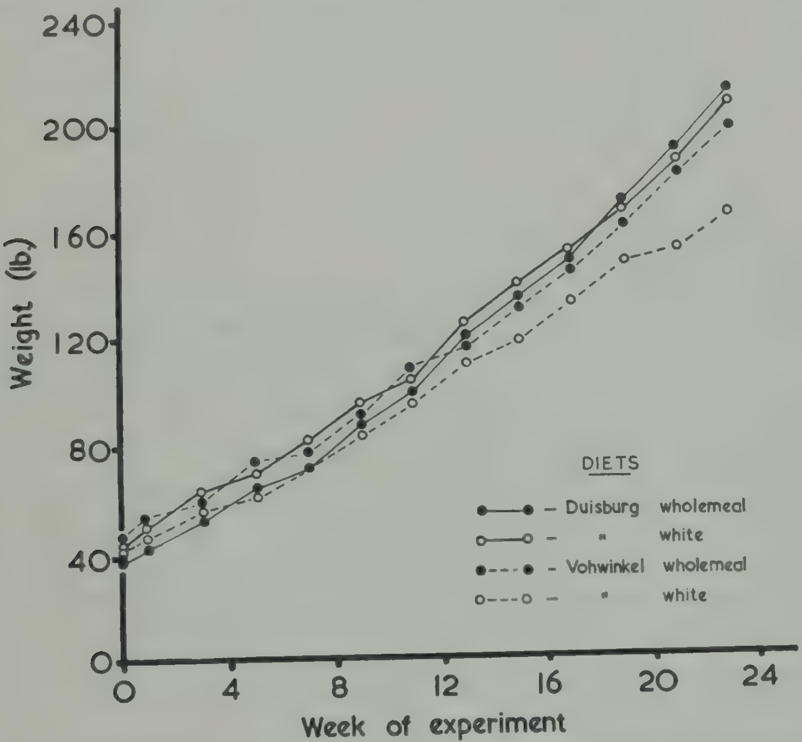

FIG. 18. Average growth rate of pigs fed on the experimental diets from 11 weeks of age.

2 pigs which were 11 weeks old when the experiment began. There was nothing to choose between the average body weights on the Duisburg diet; the 2 Vohwinkel pigs having brown flour gained a little more weight than the 2 having white.

Carcase Measurements

The carcase measurements of these pigs showed that the Vohwinkel type of diet produced a thicker layer of subcutaneous fatty tissue, a greater percentage of fat in this tissue and a greater weight of flare fat (Table 50). Since the average weights of the pairs of animals were very similar (Fig. 18), this means that the Vohwinkel animals had less flesh and bone. It may be noted that the fat of the Vohwinkel pigs had a lower iodine value. This was due to the saturated fatty acids of the margarine passing into their tissues without being metabolized.

TABLE 50

Measurements and analysis of the fatty tissues of the pigs which had been reared from 11 weeks of age on the experimental diets

Average live weight of the pigs 200 lb.

Fatty tissue	Thickness in mm. or weight in g. of the fatty tissues		Percentage of fat in the fatty tissues		Iodine value of the extracted fat	
	D.	V.	D.	V.	D.	V.
Subcutaneous fatty tissue:						
Shoulder: outer layer ..	16 mm.	17 mm.	85·4	90·3	62·5	57·0
inner layer ..	44 mm.	49 mm.	88·4	92·0	58·1	53·8
Loin: outer layer ..	15 mm.	16 mm.	88·7	90·1	61·8	56·1
inner layer ..	38 mm.	46 mm.	90·2	91·2	57·6	53·6
Rump (av. values) ..	45 mm.	50 mm.	86·5	89·8	62·1	56·6
Flare fat	900 g.	1,580 g.	93·6	93·8	50·5	50·5

D.=Duisburg diet. V.=Vohwinkel diet.

Discussion

The results of the animal experiments may be summarized in the light of the following salient features.

1. Wholemeal flour promoted better growth in *weanling* rats than white flour. This result was in keeping with those of Chick (1940, 1942), and showed once again that, given the appropriate conditions, wholemeal flour could be demonstrated to be a superior food for growing animals.

2. Rats 8 weeks old and piglets 11 weeks old gained weight no faster on diets providing 75 per cent of their calories as wholemeal flour than they did on diets in which the wholemeal was replaced by white flour. These findings resembled those at the orphanages, and confirmed the probability of their correctness. It would appear that the beneficial effects of wholemeal flour are only apparent under certain conditions, e.g. if the tests are made on rats directly after weaning when they are growing rapidly or on pigs when the diet consists almost entirely of bread (Childs *et al.*, 1948).

3. Undernourished male rats 8 weeks old grew very rapidly and equally well on diets containing wholemeal or white flour. After the first 10 weeks of the experiment, those receiving white flour began to grow slightly better than those receiving wholemeal flour.

4. Rats reared on the Vohwinkel diets ate 10 per cent more food in terms of calories but gained weight less rapidly than those reared on the Duisburg diets. At 15 weeks the Vohwinkel animals were smaller than the Duisburg animals but contained similar proportions of fat to lean. By the time they were mature the Vohwinkel animals had become as heavy and their bodies contained more fat. Piglets reared on the Vohwinkel diets were allowed 10 per cent more calories than those being reared on the Duisburg diets. They gained weight as rapidly, but at 34 weeks their carcases contained more fat and less flesh than those of pigs reared on the Duisburg diets.

The Duisburg diet produced faster growth of the fat-free parts of the body in rats and pigs, and animals with better proportions, than the Vohwinkel diet,

and this was almost certainly the case with the children too. The children at Vohwinkel gained no more weight than those at Duisburg although they consumed about 10 per cent more food in terms of calories. The clinical assessors considered that they were fatter, and the animal findings suggest that this impression was correct. There is no doubt, however, that the Vohwinkel diet would be selected by most people as the preferable one. The advantages of the Duisburg diet would probably have become more obvious as children approached maturity and might be still more striking in adult life.

PART IV: CONCLUSIONS

THE first conclusion to be drawn from this report is unquestionably that the greatest caution must be exercised in coming to any conclusion at all. Any conclusions that may be drawn must be restricted to the setting in which the scientific evidence was obtained.

Under the particular conditions of the experiments reported here no difference could be detected between the nutritive value of the different breads except when they were given to weanling rats. Probably the most important finding concerns the high nutritive value of wheat in any of the forms customarily consumed by man. Thus it has been shown that diets in which 75 per cent of the calories were derived from wheat flour and 21 per cent from vegetables, and which contained only 8 g. of animal protein a day, provided undernourished children aged 5–15 years with all the nutrients required for a high rate of growth and development for a period of 18 months.

The addition of 500 ml. of reconstituted full-cream dried milk per day over a period of 6 months caused no apparent improvement in the growth or health of the children. It is evident that diets containing much bread and little animal protein can be made highly satisfactory, and that a balanced diet, adequate in all its nutritional aspects, can be provided with minimal amounts of milk and meat, if plenty of wheat and vegetables are available.

Summary

1. The growth and development of undernourished children aged 5–15 years have been followed for a period of one year while the children were living on diets of the following composition:

Wheat flour or bread fortified with calcium carbonate	75 per cent of calories
Potatoes	6 per cent of calories
Soups, vegetables etc.	15 per cent of calories
Milk and other animal foods ..	4 per cent of calories

Supplements of vitamins A, D and C

Flours of 100 per cent, 85 per cent and 70 per cent extraction were compared and also flours of 70 per cent extraction enriched with B-vitamins and iron.

2. Under the conditions of these experiments wholemeal flour, flour of 85 per cent extraction and white flour have been found to be equally nutritious, and the enrichment of the white flour with B-vitamins and iron did not improve its nutritive value.

3. In an experiment at another orphanage, no differences were found in the nutritive value of white bread, enriched white bread and wholemeal bread when these contributed 35 per cent of the total calories and another 35 per cent were derived from fat and sugar.

4. The undernourished children who were allowed unlimited amounts of the diet listed in (1) gained in one year $1\frac{1}{2}$ times as much weight as the 'normal' growth rate of American children would have led one to expect.

5. After one year on this diet, growth was not improved by the addition of 500 ml. of reconstituted full-cream dried milk a day for a further period of 6 months.

6. Rats have been fed on diets similar in composition to those eaten by the children. Wholemeal and white flour promoted equally good growth in well-nourished or undernourished animals 8 weeks old when the experiments began; only when the rats were given the diets immediately after weaning did wholemeal flour show any superiority over white.

7. When the children's diets described in (3) were used for pigs aged 11 weeks and upwards, wholemeal flour produced a slightly more rapid increase in weight but this difference was not apparent when the flour provided 75 per cent of the total calories, as in (1).

Acknowledgements

This investigation was only made possible by the co-operation of many people both in England and in Germany and we would like to record our appreciation of all the willing help we have received. Dr. R. F. A. Dean accepted a great deal of the clinical responsibility and he and Dr. A. D. Barlow conducted the clinical examinations. Miss M. Spargo, Miss R. Selley and Miss E. Williamson supervised the food and meals at the orphanages every day and Miss L. A. Thrussell stepped into the breach on many occasions. We would also like to thank Mrs. M. Sitnik for the able way in which she made the many calculations involved in preparing the results of these experiments for publication.

We are greatly indebted to Dr. Müller-Voigt and Dr. Gehrt for allowing us to make the investigations on the children in their charge, and to Herr and Frau Hardenberg, Frau Büchsenschütz and their assistants without whose whole-hearted co-operation no work at the orphanages could have been a success.

References

Aykroyd, W. R. and Krishnan, B. G. (1939). A further experiment on the value of calcium lactate for Indian children. *Indian J. med. Res.*, **27**, 409.

Berridge, F. R. (1951). Studies of undernutrition, Wuppertal 1946–9. 4. Radiological observations on the alimentary tract. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

Berridge, F. R. and Prior, K. M. (1952). Radiological studies of the alimentary tracts of undernourished German children. *Brit. J. Radiol.*, **25**, 145.

Chick, H. (1940). Nutritive value of white flour with vitamin B_1 added and of wholemeal flour. *Lancet*, **2**, 511.

Chick, H. (1942). Biological value of the proteins contained in wheat flours. *Lancet*, **1**, 405.

Childs, G. A., Macrae, T. F., Braude, R. and Kon, S. K. (1948). Nutritive value for pigs of breads made from flours of 70, 80 and 85% extraction and effect of supplementation with aneurin, riboflavin and nicotinamide. *Brit. J. Nutr.*, **2**, ii.

Clark, M. L. (1929). Milk consumption and the growth of schoolchildren. *Lancet*, **1**, 1270.

Corry Mann, H. C. (1926). Diets for boys during the school age. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 105.

Davis, D. R. (1951). Studies of undernutrition, Wuppertal 1946–9. 8. Emotional disturbances and behavioural reactions. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

Dean, R. F. A. (1953). Plant proteins in child feeding. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 279.

Engel, C., Gorter, A. and Van Eekelen, M. (1951). De voedingswaarde van brood. IV. Groeiproeven met ratten op een gemiddelk Neederlands dieet met verschillende soorten brood. *Voeding*, **12**, 441.

Fysh, C. F. (1950). Haemoglobin determinations of 1265 Bunbury school children and of a small group of adults. *Med. J. Aust.*, **2**, 508.

Hutchinson, A. O., McCance, R. A. and Widdowson, E. M. (1951). Studies of undernutrition, Wuppertal 1946–9. 15. Serum cholinesterases. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

Hutchinson, A. O. and Widdowson, E. M. (1952). Cholinesterase activities in the serum of healthy British children. *Nature*, **169**, 284.

Leighton, G. and Clark, M. L. (1929). Milk consumption and the growth of school-children. *Lancet*, **1**, 40.

Loewenthal, L. J. A. (1938). The effect of the addition of milk to the diet of school-boys in Buganda. *E. Afr. med. J.*, **15**, 35.

McCance, R. A. and Barrett, A. M. (1951). Studies of undernutrition, Wuppertal 1946–9. 3. The effect of undernutrition on the skin. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

McCance, R. A. and Dean, R. F. A. (1951). Studies of undernutrition, Wuppertal 1946–9. 7. Neuromuscular system: Tendon reflexes and galvanic responses. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

McCance, R. A. and Walsham, C. M. (1948). The digestibility and absorption of the calories, proteins, purines, fat and calcium in wholemeal wheaten bread. *Brit. J. Nutr.*, **2**, 26.

McCance, R. A. and Widdowson, E. M. (1942). Mineral metabolism of healthy adults on white and brown bread dietaries. *J. Physiol.*, **101**, 44.

McCance, R. A. and Widdowson, E. M. (1946). The chemical composition of foods. 2nd Ed. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 235.

McCance, R. A. and Widdowson, E. M. (1951a). Studies of undernutrition, Wuppertal 1946–9. 1. The German background. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

McCance, R. A. and Widdowson, E. M. (1951b). A method of breaking down the body weights of living persons into terms of extracellular fluid, cell mass and fat, and some applications of it to physiology and medicine. *Proc. Roy. Soc., B*, **138**, 115.

McCance, R. A., Widdowson, E. M. and Shackleton, L. R. B. (1936). The nutritive value of fruits, vegetables and nuts. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 213.

Medical Research Council's Accessory Food Factors Committee (1940). M.R.C. Memorandum on bread. *Lancet*, **2**, 143, *or* Improved quality of bread. *Brit. med. J.*, **2**, 164.

Medical Research Council's Committee on Haemoglobin Surveys (1945). Haemoglobin levels in Great Britain in 1943 (with observations upon serum protein levels). *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 252.

Mellanby, M. and Coumoulos, H. (1944). The improved dentition of 5-year old London school children. Comparison between 1943 and 1929. *Brit. med. J.*, **1**, 837.

Mellanby, M. and Coumoulos, H. (1946). Teeth of 5-year old London school children (Second study). Comparison between 1929, 1943 and 1945. *Brit. med. J.*, **2**, 565.

Mellanby, M. and Mellanby, H. (1948). The reduction in dental caries in 5-year old London school children (1929–1947). *Brit. med. J.*, **2**, 409.

Milk Nutrition Committee (1939). *Milk and nutrition.* Part IV. The effects of dietary supplements of pasteurised and raw milk on the growth and health of school children (final report); Summary of all researches carried out by the committee and practical conclusions. National Institute for Research in Dairying, Reading, England.

Ministry of Food (1945). Report of the conference on the post-war loaf. H.M. Stationery Office, London.

Moran, T. and Pace, J. (1942). Digestibility of high extraction wheat meals. *Nature*, **150**, 224.

Murray, M. M., Ryle, J. A., Simpson, B. W. and Wilson, D. C. (1948). Thyroid enlargement and other changes related to the mineral content of drinking water (with a note on goitre prophylaxis). *M.R.C. Memorandum* No. 18.

National Research Council Food and Nutrition Board (1948). Recommended dietary allowances. *Repr. nat. Res. Coun., Wash.*, No. 129.

O'Brien, R., Girshick, M. A. and Hunt, E. P. (1941). Body measurements of American boys and girls for garment and pattern construction. *Misc. Publ. U.S. Dep. Agric.*, No. 366. Washington, D.C.

Orr, J. B. (1928). Milk consumption and the growth of school-children. *Lancet*, **1**, 202.

Reith, J. F., Gorter, A. and Van Eekelen, M. (1949). De voedingswaarde van brood. II. Berekeningen betreffende de betekenis van verschillende soorten brood in het Nederlandse dieet. III. Berekeningen betreffende het aminozuurgehalte van het Nederlandse dieet mit verschillende soorten brood. *Voeding*, **10**, 206, 244.

Roberts, L. J., Blair, R., Lenning, B. and Scott, M. (1938). Effect of a milk supplement on the physical status of institutional children. 1. Growth in height and weight. *Amer. J. Dis. Child.*, **56**, 287.

Scharff, J. W. and Sinnadorai, S. (1937). Milk for the school child. *Malayan med. J.*, **12**, 18.

Special Report (1951). Studies of undernutrition, Wuppertal 1946–9. Appendix: Laboratory techniques. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

Spray, C. M. and Widdowson, E. M. (1951). The effect of growth and development on the composition of mammals. *Brit. J. Nutr.*, **4**, 332.

Turbott, H. B. and Rolland, A. F. (1932). The nutritional value of milk. Experimental evidence from Maori school-children. *N.Z. med. J.*, **31**, 109.

Van Eekelen, M. (1949). De voedingswaarde van brood in het kader van het totale dieet. *Landbouwk. Tijdschr.*, **61**, 711.

Walley, J. K. (1952). The plane of nutrition and other factors which influence the composition of the fatty tissues laid down in the pig. *Thesis: Cambridge University.*

Widdowson, E. M. (1947). A study of individual children's diets. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 257.

APPENDIX A: A STATISTICAL ANALYSIS OF THE GAINS IN HEIGHT AND WEIGHT

by J. O. Irwin

THE results presented in Part I of this report, which found no significant difference in the effect of the different diets, might, it seemed, be criticized on three grounds.

(i) The withdrawal of children during the course of the experiment may have been selective and so masked real effects.

(ii) The expression of the heights and weights as percentages of an American standard (O'Brien, Girshick and Hunt, 1941) might have had a distorting effect.

(iii) The German children had been underfed before the start of these trials. Consequently their growth curves may not have been of normal form, and this might affect the applicability of the results found to children in Great Britain.

To meet points (i) and (ii) comparisons of actual gains in height and weight have been made. The influence of withdrawal has been examined by paired comparisons; so also has the influence of differences in initial age, height and weight on the mean gains in height and weight for the different diet groups.

To meet point (iii) the growth curves for the children on each diet have been obtained and compared with that given by the American standard.

PAIRED COMPARISON, TO TEST WHETHER THE WITHDRAWAL OF CHILDREN DURING THE COURSE OF THE EXPERIMENT MAY HAVE BEEN SELECTIVE

Each set of two results for children initially paired, one on the 100 per cent extraction flour and the other on the 70 per cent extraction flour, was examined. The date of withdrawal of the first member of the pair to withdraw was noted. The gains in height and weight up to that time were obtained and differenced. The difference was then expressed in cm. per annum for height and kg. per annum for weight. Table 1 shows these results. Table 2 shows the results for a similar comparison between the 85 per cent and 70 per cent extraction flours. (The 85 per cent flour was used at Duisburg only.)

No group indicates any significant differences between the diets. There is a very slight overall advantage to the 70 per cent extraction flour against the 100 per cent.

If the rate of gain were related to time of withdrawal, and withdrawal itself had some relation to diet, it is conceivable that these results might be biased. To test this point, the results have been divided into three groups and mean values have been calculated for:

(1) The group in which the first member to withdraw was on bread (a) (100 per cent or 85 per cent).

(2) The group in which the first member to withdraw was on bread (b) (70 per cent).

(3) The group in which both members continued for the whole year.

TABLE 1

Rate of gain in height or weight on 70 per cent extraction flour less rate of gain on 100 per cent extraction flour in paired children up to the time of withdrawal of the first child to withdraw (expressed in cm. or kg. per annum)

Duisburg boys			Duisburg girls			Vohwinkel boys			Vohwinkel girls		
Weeks from start	Height	Weight	Weeks from start	Height	Weight	Weeks from start	Height	Weight	Weeks from start	Height	We[illegible]
26	3·2	3·2	52	7·2	0·5	8	0	4·6	8	0	[illegible]
34	1·2	−8·7	26	2·8	−4·2	14	10·4	14·5	18	7·2	−[illegible]
52	−1·1	0·9	26	−6·0	−8·6	26	−1·0	6·2	44	−3·5	−[illegible]
40	0·9	6·5	52	−3·3	−1·3	30	−5·4	−9·7	16	−2·6	[illegible]
52	0·3	−1·3	52	−0·1	−1·1	26	−1·0	14·4	10	1·6	12[illegible]
26	−0·8	−0·8	52	1·6	4·9	52	−1·7	−0·9	2	15·6	26[illegible]
40	−1·2	0·8	52	0·9	2·1	12	2·2	11·7	34	4·9	[illegible]
52	−0·2	−0·1	26	0·6	12·6	26	1·4	2·4	52	0·3	−2[illegible]
32	−0·5	−1·1	52	−0·5	−1·4	10	2·1	7·8	4	−5·2	3[illegible]
2	2·6	2·6	52	−1·1	−0·5	2	7·8	2·6	30	1·7	6[illegible]
40	0·4	−0·4	40	−0·3	−0·4	20	2·1	6·5	26	4·4	−6[illegible]
52	1·4	0·4	52	−2·1	0·1	8	14·3	−16·9	52	1·6	−1[illegible]
20	−4·4	0·3	52	2·8	1·5	52	−1·3	−2·6	48	−2·0	−1[illegible]
52	0·3	0·3	14	4·1	−3·7	10	4·2	13·5	26	2·4	2[illegible]
40	−2·3	0·5				8	2·0	13·0	52	1·8	2[illegible]
52	0·8	−0·2				20	−2·1	−1·8	52	2·5	0[illegible]
20	2·1	0·8				42	0·2	1·5	20	1·0	−0[illegible]
52	4·8	2·9				26	2·4	0·4	32	−2·9	0[illegible]
						52	1·5	−1·8			
						2	−5·2	−20·8			
						26	1·0	−3·0			
						38	−2·8	−1·5			
						10	1·0	−4·7			
						12	0	0·9			
						20	3·4	−3·4			
						36	0·6	−1·4			
Number	18	18		14	14		26	26		18	18
Mean	0·42	0·37		0·47	0·04		1·39	1·21		1·60	2·47
Standard error	0·49	0·69		0·87	1·28		0·84	1·72		1·11	1·76

Table 3 shows the results of this comparison; there are no significant differences in the mean values of the three groups. There is no evidence here, therefore, of bias due to selective withdrawal.

TABLE 2

Rate of gain in height or weight on 70 per cent extraction flour less rate of gain on 85 per cent extraction flour in paired children up to the time of withdrawal of the first child to withdraw (expressed in cm. or kg. per annum)

Duisburg boys			Duisburg girls		
Weeks from start	Height	Weight	Weeks from start	Height	Weight
26	2·4	1·0	40	3·0	0·4
52	−2·2	1·1	26	4·4	3·2
28	1·7	−1·1	4	−6·5	5·2
40	1·3	7·3	10	−1·6	5·2
52	0·1	−1·8	52	−1·1	−1·1
26	−4·2	−2·8	52	−0·3	3·8
40	−0·7	−0·4	52	−1·7	−1·5
52	−0·4	−2·5	52	−0·1	2·6
32	−4·7	−2·4	52	−0·2	−0·6
2	0·0	5·2	52	0·9	0·9
40	−0·3	2·3	40	−0·4	−1·7
52	1·1	−0·1	52	−0·9	−0·5
20	−4·2	1·8	52	3·9	1·9
52	−0·6	0·3	52	0·0	0·4
40	0·5	1·7			
52	0·3	0·0			
16	0·0	−2·3			
52	1·0	1·6			
Number	18	18		14	14
Mean ..	−0·49	0·49		−0·04	1·30
Standard error ..	0·48	0·63		0·72	0·64

POSSIBLE EFFECT OF DIFFERENCES IN INITIAL AGE, HEIGHT AND WEIGHT

Tables 4 and 5 give the mean gains in height and weight of the children who still remained in the trial 12, 24, 36 and 52 weeks from the start. The means of the different diet groups differ less than would be expected in randomly selected groups of children on the same diet.* The differences are small, in no way consistent and may justifiably be regarded as due to chance.

To make certain that these mean values were not biased by differences in mean initial ages, heights or weights between the different groups, these variables were also examined. Their mean values are shown in Tables 6, 7 and 8. For the different diet groups these values are very close to one another and, on

* This was ascertained by analyses of variance between and within diet groups; it is a consequence of the original matching of the children on different diets.

TABLE 3

Mean difference in rates of gain in height and weight of paired children on two diets (expressed in cm. or kg. per annum)

Group	70 per cent extraction flour −100 per cent extraction flour			70 per cent extraction flour −85 per cent extraction flour		
	Both members continued 52 weeks	Member on 100 per cent flour withdrew first	Member on 70 per cent flour withdrew first	Both members continued 52 weeks	Member on 85 per cent flour withdrew first	Member [illegible] per cent [illegible] withdrew [illegible]
Duisburg						
Height						
Boys ..	+0·90±0·80 (7)*	+1·23±1·22 (3)	−0·31±0·75 (8)	−0·10±0·78 (7)	+0·47±1·19 (3)	−1·20±0·[illegible]
Girls ..	+0·60±1·11 (9)	+2·35±2·36 (2)	−1·17±1·93 (3)	+0·06±0·88 (9)	−1·70±1·52 (3)	+2·20±1·[illegible]
Weight						
Boys ..	+0·41±1·01 (7)	−2·77±1·55 (3)	+1·50±0·95 (8)	−0·20±1·02 (7)	−0·37±1·56 (3)	+1·42±0·[illegible]
Girls ..	+0·53±1·42 (9)	+4·45±3·02 (2)	−4·40±2·46 (3)	+0·66±0·73 (9)	+3·60±1·27 (3)	+0·75±1·[illegible]
Vohwinkel						
Height						
Boys ..	−0·50±2·53 (3)	+2·08±1·32 (11)	+1·23±1·26 (12)			
Girls ..	+1·55±2·22 (4)	+3·11±1·41 (10)	−2·12±2·22 (4)			
Weight						
Boys ..	−1·77±4·70 (3)	−2·56±2·46 (11)	+5·41±2·35 (12)			
Girls ..	−0·48±3·88 (4)	+3·04±2·45 (10)	+4·00±3·88 (4)			

* ± indicates standard error, figures in brackets denote number in groups.

account of the original matching, differ less than would the mean values of randomly selected groups of children. Thus no correction is necessary to the values in Tables 4 and 5 on account of any differences between the means of the initial ages, heights or weights.

However, it is conceivable that the mean values in Tables 4 and 5 might still be biased if *within* the different diet groups there were a steep increase (regression coefficient) of gain in height (or weight) with initial height (or weight). For this reason regression coefficients within diet groups were calculated for height and weight. Tables 9 and 10 show the results. The regression coefficients for

TABLE 4

Mean gains in cm. of height of children in different diet groups, 12, 24, 36 *and* 52 *weeks from start*

Extraction rate of flour (per cent)	Duisburg boys				Duisburg girls			
	12 weeks	24 weeks	36 weeks	52 weeks	12 weeks	24 weeks	36 weeks	52 weeks
100	1·93 (18)*	3·24 (17)	4·71 (15)	6·81 (13)	2·51 (14)	3·88 (13)	5·38 (12)	7·33 (11)
85	1·93 (18)	3·64 (17)	5·38 (16)	7·63 (15)	1·99 (12)	3·57 (12)	4·98 (12)	7·19 (11)
70	1·75 (17)	3·45 (16)	4·94 (12)	7·38 (9)	2·31 (14)	3·96 (14)	5·54 (12)	7·69 (11)
70 enriched to 100	1·80 (17)	3·36 (16)	4·91 (14)	6·64 (9)	2·11 (14)	3·62 (14)	5·32 (12)	7·41 (8)
70 enriched to 85	1·97 (17)	3·50 (17)	5·14 (14)	7·45 (11)	1·89 (12)	3·26 (10)	4·67 (10)	7·62 (5)
	Vohwinkel boys				Vohwinkel girls			
100	1·83 (21)	3·66 (17)	5·17 (15)	7·63 (9)	1·74 (17)	3·38 (14)	5·67 (10)	7·30 (7)
70	1·97 (22)	3·55 (19)	5·10 (14)	6·94 (9)	2·04 (16)	3·87 (14)	5·91 (13)	7·83 (10)
70 enriched to 100	1·88 (22)	3·86 (18)	5·86 (14)	8·10 (11)	1·67 (16)	3·44 (16)	5·21 (11)	7·13 (9)

* Figures in brackets denote number of children in groups.

TABLE 5

Mean gains in kg. of weight of children in different diet groups, 12, 24, 36 *and* 52 *weeks from start*

Extraction rate of flour (per cent)	Duisburg boys				Duisburg girls			
	12 weeks	24 weeks	36 weeks	52 weeks	12 weeks	24 weeks	36 weeks	52 weeks
100	1·25 (18)*	2·45 (17)	2·95 (15)	3·55 (13)	1·81 (14)	3·42 (13)	4·64 (12)	5·34 (11)
85	1·47 (18)	2·34 (17)	3·18 (16)	3·99 (15)	1·50 (12)	2·52 (12)	3·58 (12)	4·32 (11)
70	1·53 (17)	2·19 (16)	3·73 (12)	4·16 (9)	2·04 (14)	3·23 (14)	4·41 (12)	5·58 (11)
70 enriched to 100	1·22 (17)	2·21 (16)	3·61 (14)	3·74 (9)	2·08 (14)	3·36 (14)	4·92 (12)	5·65 (8)
70 enriched to 85	1·09 (17)	1·85 (17)	3·31 (14)	4·44 (11)	1·98 (12)	3·45 (10)	5·06 (10)	5·90 (5)
	Vohwinkel boys				Vohwinkel girls			
100	1·44 (21)	2·65 (17)	3·36 (15)	5·54 (9)	2·15 (17)	2·89 (14)	3·51 (10)	4·16 (7)
70	1·96 (22)	2·34 (19)	2·59 (14)	3·67 (9)	2·66 (16)	3·15 (14)	3·96 (13)	4·93 (10)
70 enriched to 100	2·00 (22)	2·84 (18)	4·11 (14)	5·10 (11)	2·20 (16)	3·25 (16)	3·39 (11)	4·17 (9)

* Figures in brackets denote number of children in groups.

TABLE 6

Mean ages in years and months, at beginning of trial, of children remaining in different diet groups, 12, 24, 36 *and* 52 *weeks from start*

Extraction rate of flour (per cent)	Duisburg boys				Duisburg girls			
	12 weeks	24 weeks	36 weeks	52 weeks	12 weeks	24 weeks	36 weeks	52 weeks
100	9–9 (18)*	9–11 (17)	9–9 (15)	9–6 (13)	9–11 (14)	10–3 (13)	10–4 (12)	10–0 (11)
85	9–4 (18)	9–7 (17)	9–4 (16)	9–6 (15)	10–0 (12)	10–0 (12)	10–0 (12)	9–7 (11)
70	9–6 (17)	9–7 (16)	9–5 (12)	9–3 (9)	10–3 (14)	10–3 (14)	9–9 (12)	10–0 (11)
70 enriched to 100	9–3 (17)	9–8 (16)	9–6 (14)	8–4 (9)	10–0 (14)	10–0 (14)	10–4 (12)	10–9 (8)
70 enriched to 85	9–4 (17)	9–4 (17)	9–8 (14)	9–6 (11)	9–3 (12)	10–1 (10)	10–1 (10)	9–4 (5)
	Vohwinkel boys				Vohwinkel girls			
100	9–9 (21)	10–3 (17)	10–3 (15)	11–1 (9)	9–6 (17)	9–4 (14)	9–3 (10)	9–3 (7)
70	10–2 (22)	9–5 (19)	9–5 (14)	10–3 (9)	9–7 (16)	9–2 (14)	9–6 (13)	10–0 (10)
70 enriched to 100	9–10 (22)	9–11 (18)	10–5 (14)	10–9 (11)	9–4 (16)	9–4 (16)	8–7 (11)	9–3 (9)

* Figures in brackets denote number of children in groups.

TABLE 7

Mean heights in cm. at beginning of trial of children remaining in different diet groups, 12, 24, 36 *and* 52 *weeks from start*

Extraction rate of flour (per cent)	Duisburg boys				Duisburg girls			
	12 weeks	24 weeks	36 weeks	52 weeks	12 weeks	24 weeks	36 weeks	52 weeks
100	130·2 (18)*	131·2 (17)	129·2 (15)	128·4 (13)	127·4 (14)	129·4 (13)	130·4 (12)	129·1 (11)
85	126·4 (18)	127·0 (17)	126·0 (16)	126·1 (15)	129·3 (12)	129·3 (12)	129·3 (12)	127·3 (11)
70	126·5 (17)	126·6 (16)	126·7 (12)	125·1 (9)	131·2 (14)	131·2 (14)	129·5 (12)	131·3 (11)
70 enriched to 100	127·3 (17)	126·7 (16)	126·0 (14)	120·9 (9)	128·5 (14)	128·5 (14)	131·9 (12)	132·4 (8)
70 enriched to 85	126·7 (17)	126·7 (17)	128·2 (14)	126·9 (11)	127·6 (12)	131·4 (10)	131·4 (10)	127·3 (5)
	Vohwinkel boys				Vohwinkel girls			
100	129·0 (21)	130·5 (17)	130·4 (15)	136·8 (9)	128·3 (17)	127·0 (14)	127·1 (10)	128·1 (7)
70	130·4 (22)	128·5 (19)	125·2 (14)	128·2 (9)	128·1 (16)	125·8 (14)	127·8 (13)	129·7 (10)
70 enriched to 100	131·0 (22)	131·0 (18)	132·1 (14)	133·4 (11)	127·8 (16)	127·8 (16)	123·5 (11)	126·7 (9)

* Figures in brackets denote number of children in groups.

TABLE 8

Mean weights in kg. at beginning of trial of children remaining in different diet groups, 12, 24, 36 and 52 weeks from start

Extraction rate of flour (per cent)	Duisburg boys 12 weeks	Duisburg boys 24 weeks	Duisburg boys 36 weeks	Duisburg boys 52 weeks	Duisburg girls 12 weeks	Duisburg girls 24 weeks	Duisburg girls 36 weeks	Duisburg girls 52 weeks
100	29·1 (18)*	29·6 (17)	28·0 (15)	27·5 (13)	29·0 (14)	29·8 (13)	30·5 (12)	29·5 (11)
85	27·1 (18)	27·3 (17)	27·2 (16)	27·2 (15)	28·6 (12)	28·6 (12)	28·6 (12)	27·4 (11)
70	27·0 (17)	27·1 (16)	27·2 (12)	26·5 (9)	30·8 (14)	30·8 (14)	29·5 (12)	30·3 (11)
70 enriched to 100	27·5 (17)	27·0 (16)	26·7 (14)	23·9 (9)	28·4 (14)	28·4 (14)	29·6 (12)	29·4 (8)
70 enriched to 85	27·3 (17)	27·3 (17)	28·0 (14)	27·4 (11)	29·0 (12)	30·9 (10)	30·9 (10)	28·5 (5)
	Vohwinkel boys				Vohwinkel girls			
100	28·1 (21)	28·9 (17)	29·1 (15)	32·6 (9)	27·3 (17)	26·1 (14)	26·3 (10)	26·5 (7)
70	29·4 (22)	28·4 (19)	26·6 (14)	27·9 (9)	28·3 (16)	26·9 (14)	27·7 (13)	29·0 (10)
70 enriched to 100	28·9 (22)	28·9 (18)	29·7 (14)	30·1 (11)	27·5 (16)	27·5 (16)	25·5 (11)	26·8 (9)

* Figures in brackets denote number of children in groups.

TABLE 9

Regression coefficients within diet groups of gain in height on initial height

Time from start (weeks)	Duisburg boys	Duisburg girls
12	$-0·007 \pm 0·004$	$-0·009 \pm 0·006$
24	$-0·004 \pm 0·006$	$-0·019 \pm 0·009$
36	$0·000 \pm 0·009$	$-0·030 \pm 0·013$
52	$0·010 \pm 0·014$	$-0·029 \pm 0·019$
	Vohwinkel boys	Vohwinkel girls
12	$0·001 \pm 0·006$	$-0·016 \pm 0·020$
24	$-0·007 \pm 0·008$	$-0·028 \pm 0·021$
36	$-0·007 \pm 0·013$	$-0·046 \pm 0·020$
52	$0·017 \pm 0·023$	$-0·069 \pm 0·026$

TABLE 10

Regression coefficients within diet groups of gain in weight on initial weight

Time from start (weeks)	Duisburg boys	Duisburg girls
12	$0·062 \pm 0·013$	$0·073 \pm 0·012$
24	$0·104 \pm 0·017$	$0·127 \pm 0·017$
36	$0·157 \pm 0·017$	$0·180 \pm 0·021$
52	$0·128 \pm 0·024$	$0·213 \pm 0·029$
	Vohwinkel boys	Vohwinkel girls
12	$0·118 \pm 0·022$	$0·159 \pm 0·027$
24	$0·098 \pm 0·028$	$0·150 \pm 0·033$
36	$0·130 \pm 0·029$	$0·145 \pm 0·031$
52	$0·216 \pm 0·043$	$0·137 \pm 0·044$

height are very small and in all except two cases insignificant. Those for weight are larger and statistically significant, but they would lead to quite negligible alterations in the mean values of Table 5.* Nevertheless, for weight the analysis of variance of the corrected values was carried out; it led to the same conclusion as that for the uncorrected values.†

THE ABSOLUTE GROWTH CURVES COMPARED WITH THE AMERICAN STANDARD

For each diet group, treating boys and girls at Duisburg and Vohwinkel separately, absolute growth curves were obtained. First the mean gains in height or weight at the end of each fortnight from the beginning were calculated for those children who still remained in the trial at that time. The mean initial ages, heights and weights of the same children were then similarly calculated. Then

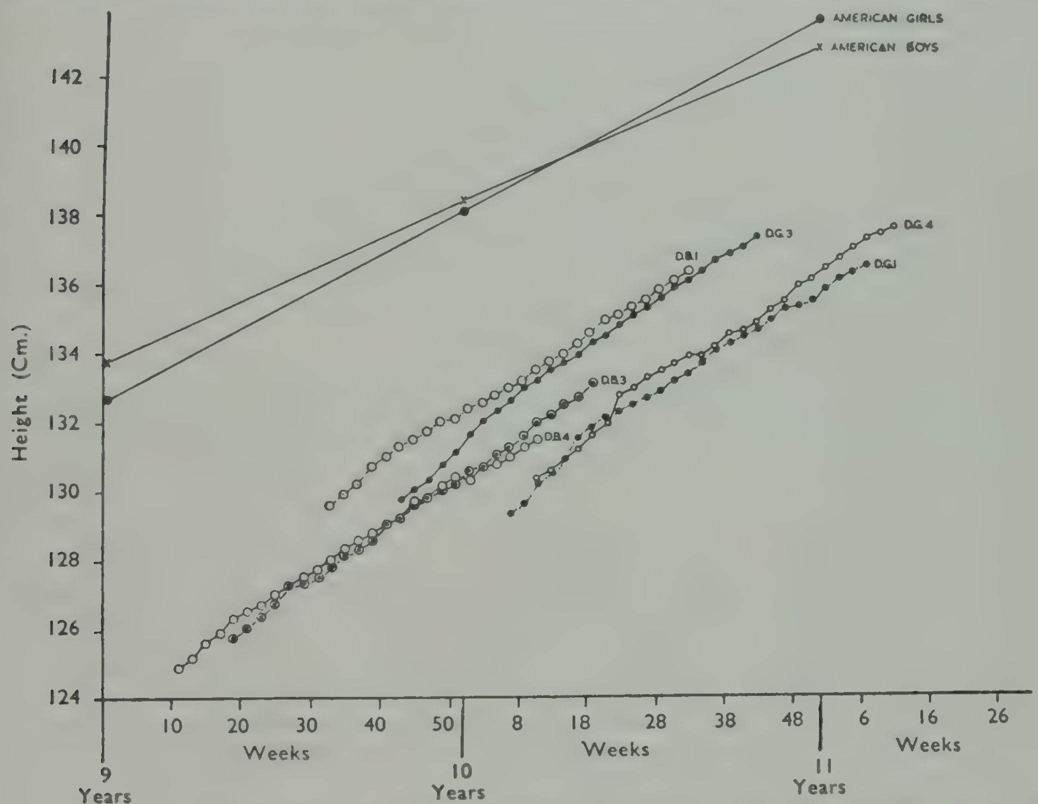

FIG. 1A. The absolute growth in height of some of the children at Duisburg compared with the American standard. D.B. = Duisburg boys, D.G. = Duisburg girls, 1 = flour of 100% extraction, 3 = flour of 70 % extraction, 4 = flour of 70% extraction enriched to 100%.

weighted means of these results were obtained.‡ These were taken as the mean initial ages, heights and weights for the group. The gains in height and weight could then be added to the initial values to complete the construction of the curves.

---

* The usual technique of pooling sums of squares and products of deviations from the individual diet groups was used.

† For example, to take an extreme case, the mean value of the gain in weight at 52 weeks for all diet groups in the Duisburg boys is 3·96 kg. The smallest value is 3·55 (100 per cent extraction). This would be corrected to 3·55 + 0·128 (0·44) = 3·61. For the corresponding groups of girls the mean for all diet groups is 5·03 kg. The lowest value is 4·32 (85 per cent extraction). This would be altered to 4·32 + 0·212 (0·71) = 4·46.

‡ Actually, the fortnightly means were weighted by the product of the time from the start and the number of children. Almost identical results could have been obtained from unweighted means, or weighting by number of children only.

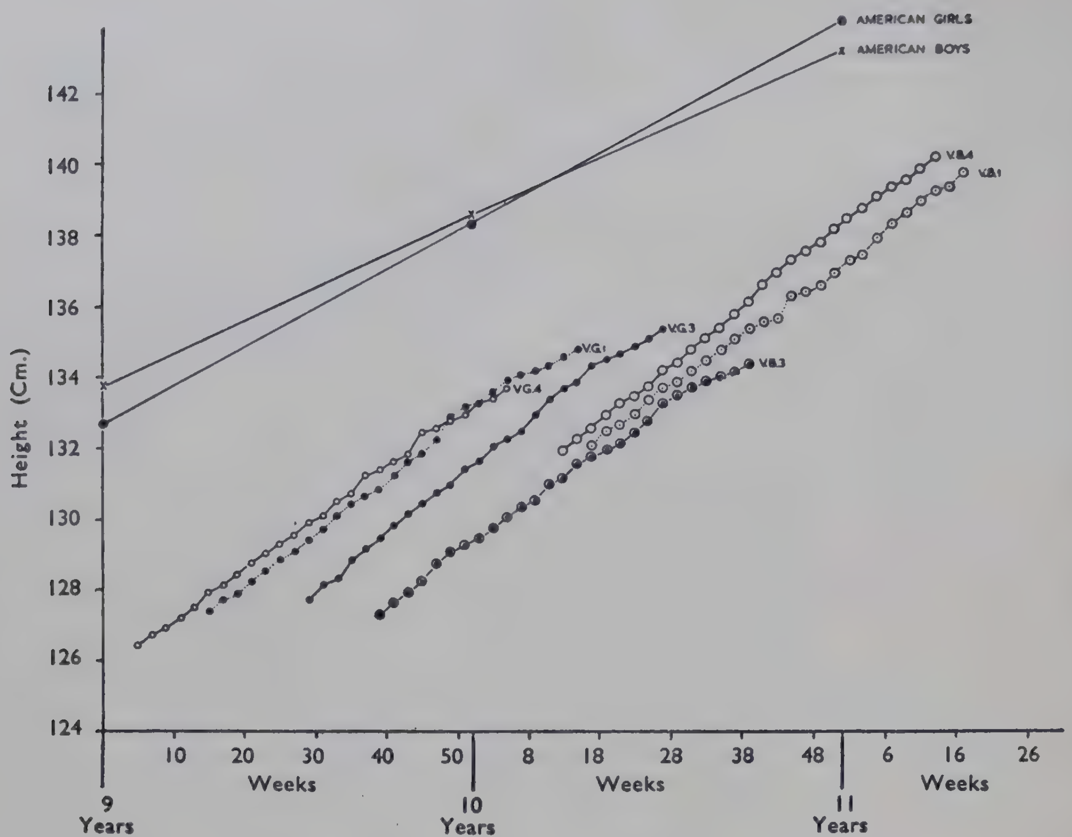

FIG. 1B. The absolute growth in height of the children at Vohwinkel compared with the American standard. V.B. = Vohwinkel boys, V.G. = Vohwinkel girls, 1, 3, 4 as in Fig. 1A.

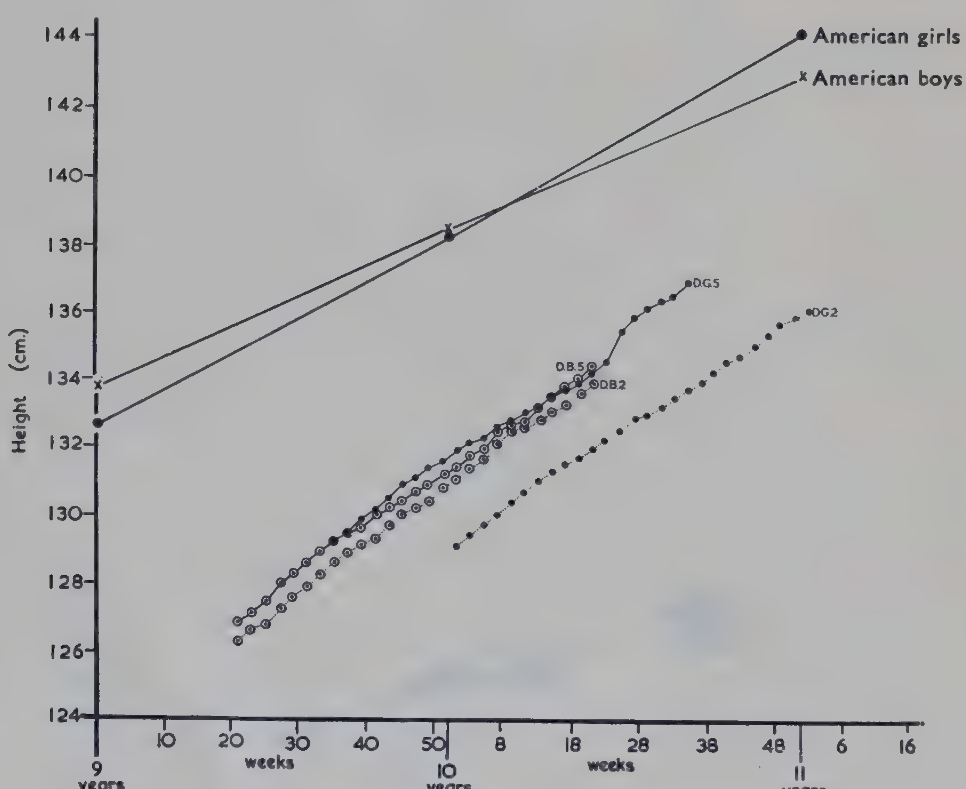

FIG. 1C. The absolute growth in height of the other children at Duisburg compared with the American standard. D.B. = Duisburg boys, D.G. = Duisburg girls, 2 = flour of 85% extraction, 5 = flour of 70% extraction enriched to 85%.

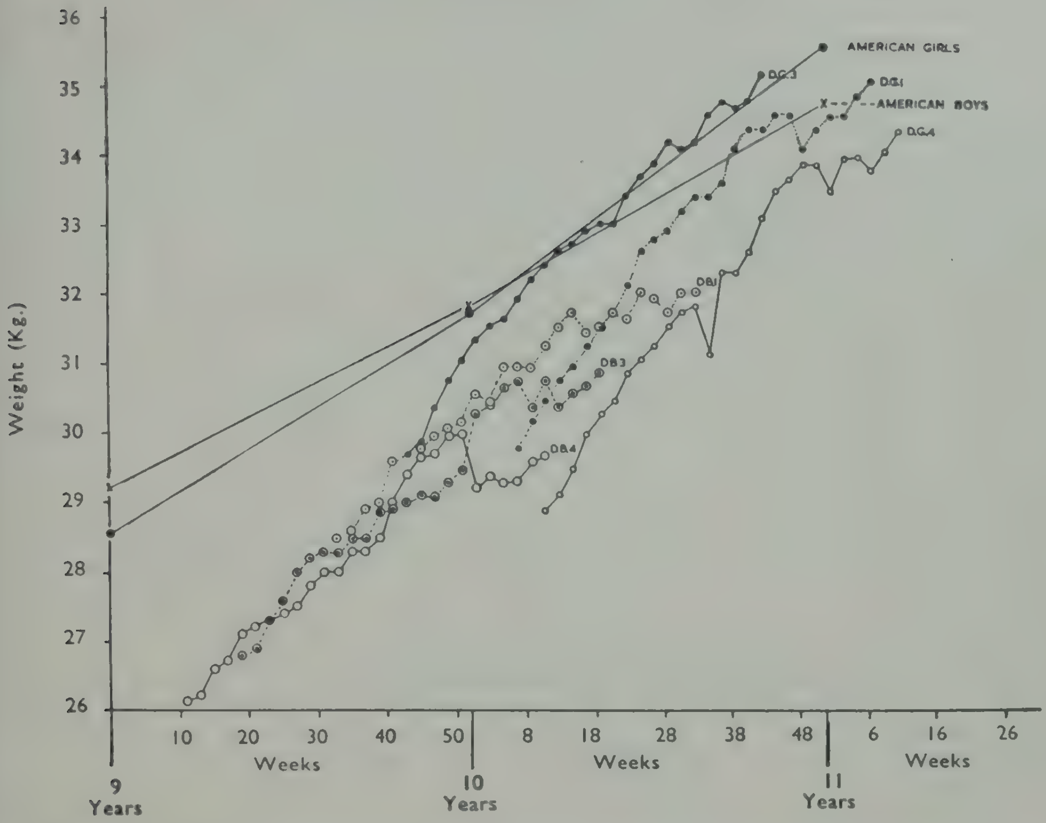

FIG. 2A. The absolute growth in weight of some of the children at Duisburg compared with the American standard. D.B. = Duisburg boys, D.G. = Duisburg girls, 1 = flour of 100% extraction, 3 = flour of 70% extraction, 4 = flour of 70% extraction enriched to 100%.

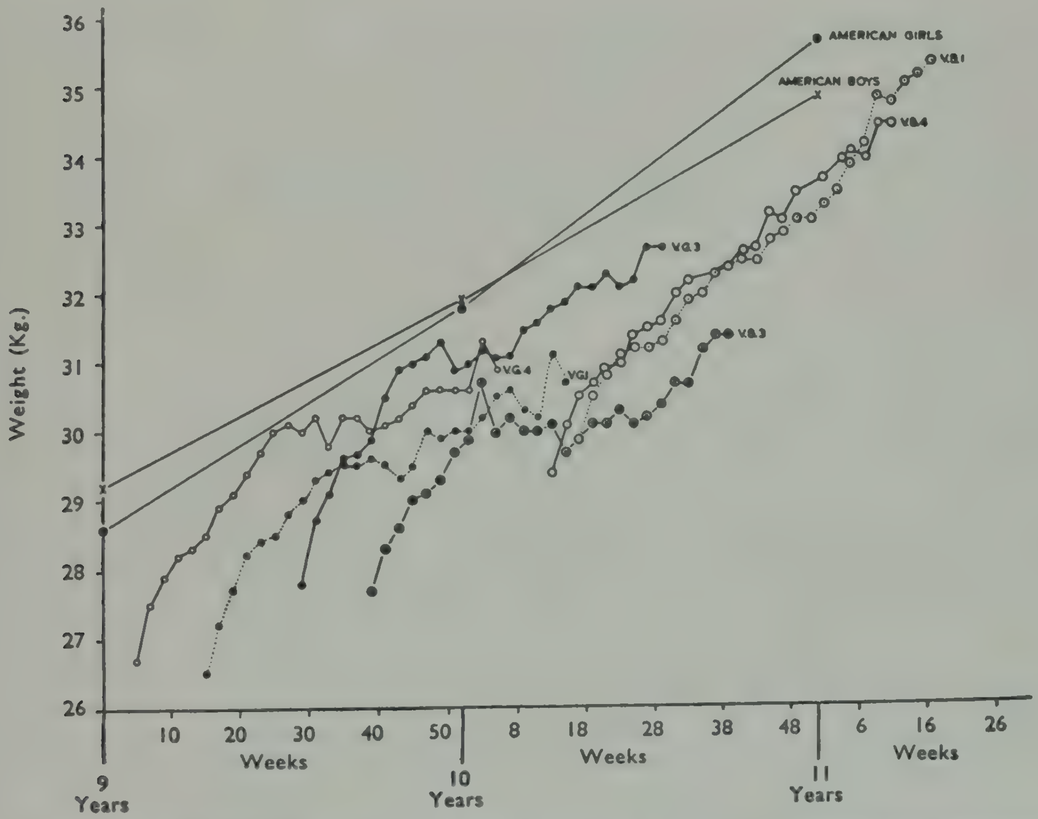

Fig. 2B. The absolute growth in weight of the children at Vohwinkel compared with the American standard. V.B. = Vohwinkel boys, V.G. = Vohwinkel girls, 1, 3, 4 as in Fig. 2A.

The curves are shown in Figs. 1 A, B and C, and 2 A, B and C, where the American standard has been inserted for comparison. They must be interpreted with caution. There are no significant differences in slope or position between the different diet groups. Only mean values, of course, have been plotted; there would be considerable overlap of results for individual children of the same age in different diet groups. The curves for height appear linear and are on the

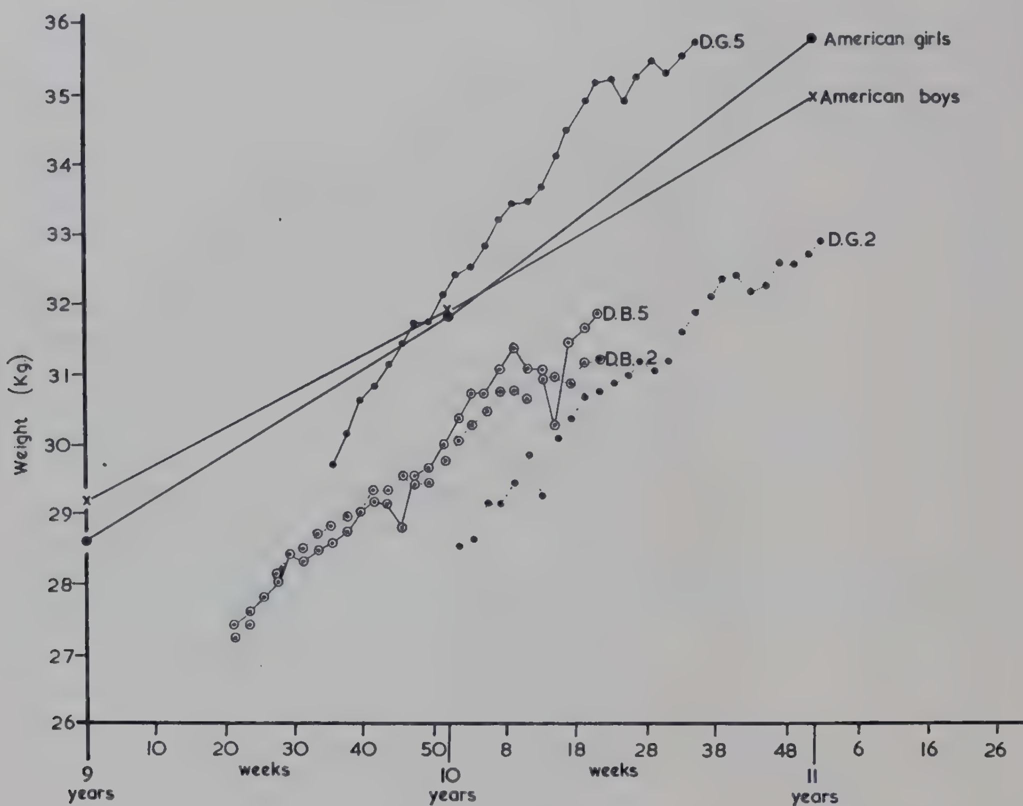

FIG. 2C. The absolute growth in weight of the other children at Duisburg compared with the American standard. D.B. = Duisburg boys, D.G. = Duisburg girls, 2 = flour of 85% extraction, 5 = flour of 70% extraction enriched to 85%.

average slightly steeper than the corresponding American standard curves. This is more noticeable in the curve for weight. In the latter the rate of increase is greater in the earlier than in the later part of the experiment.

The general level of the curves for German children is below that of the American standard. Apart from any genetic differences that may exist this would naturally be expected in children who had been previously underfed.

Doubtless one school of thought will argue, particularly from the weight curves, that, because of underfeeding, these children would have grown on any diet not grossly inadequate, that the response to different amounts of vitamins would therefore be largely non-specific, and that specific differences in response might still be detected in children with a more normal previous history kept on similar diets for a longer period of time. Others will be more struck by the fact that these children grew equally well on all the diets and at a rather faster rate than indicated by the standard curves for American children. It is not possible, from the statistical data, to decide between these alternatives.

Summary

No significant difference due to diet has been detected in the increase in height and weight of the German children who underwent this trial. There is no evidence that the results were biased by selective withdrawal of children during the course of the trial, or by initial differences of age, height or weight in the different groups. The children grew slightly faster in height and faster in weight than is indicated by an American standard. Their increase in weight was faster in the earlier part of the trial than the later. The relation of these findings to the specificity of the response to the different diets is briefly discussed. It is not possible from the statistical data to decide between the possible alternative explanations.

References

O'Brien, R., Girshick, M. A. and Hunt, E. P. (1941). Body measurements of American boys and girls for garment and pattern construction. *Misc. Publ. U.S. Dep. Agric.*, No. 366. Washington, D.C.

APPENDIX B: THE NITROGEN AND MINERAL METABOLISM OF CHILDREN EATING THE DIFFERENT EXPERIMENTAL BREADS

by E. M. Widdowson and Lois A. Thrussell

It was clearly impossible to make metabolic balance studies on all the children over the whole year of the experiments, but it was felt that some metabolic work would be of value in assessing the nutritional adequacy of the different diets. 'Sample' children were accordingly chosen from each of the bread groups at the two homes, and their intakes and excretions of nitrogen, calcium, phosphorus and magnesium, as well as of the B-vitamins (Holman, p. 92) were measured for one week. The methods used for the collection and analysis of the samples are given on p. 21.

These metabolic studies were made after the children had been living on the experimental diets for 3–7 months. Five boys and 10 girls were investigated at Duisburg and 6 boys at Vohwinkel. The ages of these children and their heights and weights at the time when these particular investigations were being made are shown in Table 1. The heights and weights are expressed as a percentage of the 'normal', as taken from the figures of O'Brien, Girshick and Hunt (1941) and described on p. 5.

Tables 2 and 3 give information about the average weight of bread, or of bread and cake, eaten by each child each day during the experimental week, and the average daily calorie intake over that week compared with the average calorie intake of the same children over the whole year. It will be seen that for nearly all the children the two figures for calories agree reasonably closely, and the diets during the week of metabolism may therefore be taken as being fairly representative of the diet over the whole year. 'Hanni', and to a lesser extent 'Helga', at Duisburg were exceptions. Both were unsettled by the metabolic experiment and did not eat their normal amounts of food. The results which were obtained on them have been included in the tables, but the averages have been calculated both with and without figures for Hanni.

The weights of faeces and volumes of urine passed during the week of metabolism are also shown in Tables 2 and 3. At Duisburg the faeces passed by the children eating the bread made from flour of 100 per cent extraction weighed on average more than twice as much as those passed by the children in any other group. Each of the 3 children having the wholemeal flour passed a greater weight of faeces than any of the other 12 children and the dry weights of these faeces were more than twice those of any of the others, so that the difference was due to a greater excretion of solids as well as of water. These findings are in accordance with previous experience (Meyer, 1871; Rubner, 1883, 1917; Snyder, 1905, 1908; Newman, Robinson, Halnan and Neville, 1912; McCance and Widdowson, 1942, 1946). The differences between the groups at Vohwinkel were similar to, but smaller than, those at Duisburg.

The Absorption and Excretion of Nitrogen

Table 4 shows the intake and output of nitrogen of the children in the different bread groups at the two homes. At Duisburg the intakes were of the order of 10–15 g. a day, which is equivalent to 60–90 g. of protein. About three quarters of this came from the bread. The children receiving bread made from flour of

TABLE 1

Description of the children taking part in the metabolic studies

Extraction rate of flour (per cent)	Name	Sex	Age (years and months)	Date of metabolic study	Height as per cent of 'normal'	Weight as per cent of 'normal'	Average gain in weight over whole year (g. per day)
				Duisburg			
100	Hilde	F	12–5	Sept. 1947	100·0	103·2	17·3
	Bernhard	M	12–10	Nov. 1947	95·7	90·8	7·7
	Erika	F	13–11	Jan. 1948	95·5	102·4	31·9
	Average		13–1		97·1	98·8	19·0
85	Helga	F	12–0	Sept. 1947	97·1	95·3	14·3
	Horst	M	11–9	Nov. 1947	90·3	88·3	14·0
	Agnes	F	15–1	Jan. 1948	97·4	87·0	22·7
	Average		12–11		94·9	90·2	17·0
70	Marianne	F	13–0	Sept. 1947	91·7	82·4	14·3
	Helmut	M	13–5	Nov. 1947	96·6	102·0	26·7
	Irmtraud	F	14–9	Jan. 1948	94·8	84·0	23·9
	Average		13–9		94·4	89·5	21·6
70 enriched to 100	Hannelore	F	13–1	Sept. 1947	98·6	97·2	25·5
	Harro	M	12–2	Nov. 1947	100·7	104·6	23·8
	Ursula	F	14–7	Jan. 1948	97·8	87·0	16·2
	Average		13–3		99·0	96·3	21·8
70 enriched to 85	Hanni	F	13–1	Sept. 1947	97·5	111·0	31·0
	Günther	M	11–6	Nov. 1947	99·7	113·4	15·4
	Frieda	F	14–11	Jan. 1948	98·5	108·8	28·2
	Average		13–2		98·6	111·1	24·9
				Vohwinkel			
100	Alvin	M	11–3	May 1948	97·2	104·1	18·7
	Willi	M	13–3	May 1948	97·4	101·8	18·7
	Average		12–3		97·3	102·9	18·7
70	Heinz	M	12–10	May 1948	98·0	103·8	14·8
	Hans	M	11–3	May 1948	97·2	97·7	18·7
	Average		12–0		97·6	100·7	16·7
70 enriched to 100	Klaus	M	13–7	May 1948	92·0	90·5	15·1
	Karl-Heinz	M	12–1	May 1948	96·5	103·9	20·3
	Average		12–10		94·3	97·2	17·7

TABLE 2

Average daily amounts of food eaten and of faeces and urine passed during the metabolic studies at Duisburg

Extraction rate of flour (per cent)	Name	Weight bread eaten (g. per day)	Average daily Calories over week of metabolic study	Average daily Calories over whole year	Weight faeces as passed (g. per day)	Dry weight faeces passed (g. per day)	Urine volume (ml. per day)
100	Hilde ..	728	2,280	2,520	320	67	1,375
	Bernhard	767	2,470	2,237	373	79	1,440
	Erika	729	2,589	2,528	355	63	1,974
	Average	741	2,446	2,428	349	70	1,596
85	Helga ..	438	1,610	2,193	99	24	1,370
	Horst ..	716	2,359	2,271	217	35	1,352
	Agnes ..	689	2,545	2,263	193	30	2,030
	Average	614	2,171	2,242	170	30	1,584
70	Marianne	537	1,926	2,278	66	18	1,288
	Helmut	763	2,562	2,479	72	22	1,694
	Irmtraud	662	2,560	2,236	134	22	2,009
	Average	654	2,349	2,331	91	21	1,664
70 enriched to 100	Hannelore	608	2,170	2,435	177	28	1,674
	Harro ..	830	2,762	2,668	213	34	1,693
	Ursula ..	560	2,293	2,292	95	21	2,399
	Average	666	2,408	2,465	162	28	1,922
70 enriched to 85	Hanni ..	385	1,517	2,521	69	21	1,330
	Günther	725	2,479	2,648	72	21	1,344
	Frieda ..	696	2,720	2,604	100	23	2,093
	Average	602	2,239	2,591	80	22	1,589
	Average, excluding 'Hanni'	710	2,600	2,626	86	22	1,718

100 per cent extraction ate more bread than the others and their nitrogen intakes were a little higher. They excreted nearly twice as much in the faeces, however, so that their apparent absorption was a smaller proportion of their intake than it was in the other groups. Previous work (McCance and Walsham, 1948) suggests that these children probably did, in fact, absorb all the nitrogen in their diets, and that the nitrogen found in their faeces was entirely metabolic in origin. There was no significant difference between the average retentions of any of the groups.

TABLE 3

Average daily amounts of foods eaten and of faeces and urine passed during the metabolic studies at Vohwinkel

Extraction rate of flour (per cent)	Name	Weight bread eaten (g. per day)	Weight cake eaten (g. per day)	Average daily Calories over week of metabolic study	Average daily Calories over whole year	Weight faeces as passed (g. per day)	Urine volume (ml. per day)
100	Alvin ..	244	219	2,841	2,885	215	1,429
	Willi ..	221	211	2,749	2,684	240	1,401
	Average	232	215	2,795	2,784	227	1,415
70	Heinz ..	226	185	2,703	3,005	207	1,220
	Hans ..	200	175	2,529	2,700	185	1,389
	Average	213	180	2,616	2,852	196	1,304
70 enriched to 100	Klaus ..	230	217	2,856	2,470	135	1,473
	Karl-Heinz	221	234	2,885	2,712	178	1,167
	Average	226	225	2,870	2,591	156	1,320

At Vohwinkel the intake of nitrogen from bread and flour was not much more than half that at Duisburg, but foods other than cereals provided considerably more nitrogen than the non-cereal part of the Duisburg diet. The nitrogen intakes at Vohwinkel were therefore only a little lower than they were at Duisburg. The Vohwinkel children absorbed a smaller percentage of their intakes, which may have been related to the general composition or to a single component of their diets. They excreted a correspondingly smaller amount in the urine, so that their retentions were very similar to those at Duisburg and amounted to about 2 g. a day for the children in all the groups.

A nitrogen retention of 2 g. a day is similar to the value found by Widdowson and Thrussell (1951) for a group of German children who were not taking part in the investigations described in this report, but were living on a similar type of diet. It is considerably higher than the figure of 0·8 g. given by Macy (1942) for the daily nitrogen retention of two well-nourished children aged 11 and 12 years whose intakes were similar to those of the Duisburg and Vohwinkel children but whose diets included 55 g. of animal protein a day.

Assuming the cell mass of the body to contain 33 per cent of protein, a daily retention of 2 g. of nitrogen is equivalent to a daily increment of 37·5 g. of cell mass. The actual gain in weight of the children who took part in the metabolic studies could not be measured accurately over a single week, but over the whole year of the experiment it averaged about 20 g. a day (Table 1). This gain included fat as well as muscle, and the results therefore suggest that the children stored more nitrogen, and probably gained more weight, during the metabolic week

TABLE 4

Absorption and excretion of nitrogen on the experimental diets

Extraction rate of flour (per cent)	Name	Intake of nitrogen (g. per day)			Excretion of nitrogen (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Excretio in urine as per cer of intak
		Bread and flour	Other foods	Total	Urine	Faeces	Total				
					Duisburg						
100	Hilde	10·8	3·3	14·1	10·5	2·6	13·1	+ 1·0	11·5	81·5	74·5
	Bernhard	11·4	3·5	14·9	8·1	4·0	12·1	+ 2·8	10·9	73·1	54·3
	Erika	11·1	4·2	15·3	10·0	2·6	12·6	+ 2·7	12·7	83·0	65·3
	Average	11·1	3·7	14·8	9·5	3·1	12·6	+ 2·2	11·7	79·1	64·2
85	Helga	6·7	3·3	10·0	8·6	1·3	9·9	+ 0·1	8·7	87·0	86·0
	Horst	10·3	3·4	13·7	9·6	2·0	11·6	+ 2·1	11·7	85·4	70·0
	Agnes	10·3	4·1	14·4	9·6	2·0	11·6	+ 2·8	12·4	86·1	66·6
	Average	9·1	3·6	12·7	9·3	1·7	11·0	+ 1·7	10·9	85·8	73·2
70	Marianne	7·5	3·3	10·8	8·1	1·3	9·4	+ 1·4	9·5	88·0	75·0
	Helmut	10·3	3·5	13·8	9·7	1·4	11·1	+ 2·7	12·4	89·8	70·3
	Irmtraud	9·7	4·1	13·8	9·4	1·8	11·2	+ 2·6	12·0	87·0	68·1
70 enriched to 100	Hannelore	8·6	4·0	12·6	9·8	1·7	11·5	+ 1·1	10·9	86·5	77·8
	Harro	11·6	3·5	15·1	10·9	2·3	13·2	+ 1·9	12·8	84·8	72·2
	Ursula	8·2	4·1	12·3	8·6	1·4	10·0	+ 2·3	10·9	88·6	69·9
70 enriched to 85	Hanni	5·8	3·3	9·1	9·2	1·3	10·5	− 1·4	7·8	85·7	101·1
	Günther	9·5	3·5	13·0	9·8	1·5	11·3	+ 1·7	11·5	88·4	75·4
	Frieda	10·4	4·1	14·5	10·1	1·5	11·6	+ 2·9	13·0	89·7	69·6
	Average all 70 per cent flours	9·1	3·7	12·8	9·5	1·6	11·1	+ 1·7	11·2	87·5	74·2
	Average excluding 'Hanni'	9·5	3·8	13·3	9·5	1·6	11·1	+ 2·2	11·7	88·0	71·4
					Vohwinkel						
100	Alvin	6·1	5·3	11·4	7·4	2·3	9·7	+ 1·7	9·1	79·8	64·9
	Willi	5·7	5·3	11·0	6·3	2·6	8·9	+ 2·1	8·4	76·3	57·3
	Average	5·9	5·3	11·2	6·8	2·5	9·3	+ 1·9	8·7	77·7	60·7
70	Heinz	5·5	5·2	10·7	7·1	1·9	9·0	+ 1·7	8·8	82·2	66·3
	Hans	5·0	5·2	10·2	5·6	2·2	7·8	+ 2·4	8·0	78·4	54·9
70 enriched to 100	Klaus	5·8	5·3	11·1	7·3	1·9	9·2	+ 1·9	9·2	82·9	65·8
	Karl-Heinz	5·8	5·3	11·1	6·7	2·0	8·7	+ 2·4	9·1	82·0	60·3
	Average all 70 per cent flours	5·5	5·3	10·8	6·7	2·0	8·7	+ 2·1	8·8	81·5	62·0

than they did in most other weeks during the year. A possible reason for this behaviour is that the extra care which they received during the metabolic study promoted nitrogen retention. The very nature of such an investigation makes it necessary to devote far more attention to the children than they would be likely to receive at other times, particularly if they are living in an orphanage, and Widdowson (1951) has shown that children who were handled sympathetically gained more weight than others who were treated harshly, even though their diets were the same. There is, however, another explanation, which can only be discussed very tentatively at the present time. It depends upon the possibility that the amount of protein in the cell is not such a fixed quantity as people have

supposed, and that it may increase or decrease within certain limits according to the age and state of nutrition. This need not be accompanied by the expected change in the weight of the body if the amount of fluid in the cell were to change in the opposite direction at the same time.

The Absorption and Excretion of Minerals

Determinations of the intake and output of calcium, phosphorus and magnesium were made on the children at Duisburg only. The detailed results are set out in Tables 5, 6 and 7.

Calcium

All the children were taking more than 1 g. of calcium a day and those having the 100 per cent extraction flour about 2·5 g. The non-cereal part of the diet provided 0·6–0·7 g. a day, which was almost as much as Widdowson (1947)

TABLE 5

Absorption and excretion of calcium on the experimental diets at Duisburg

xtraction rate of flour (per cent)	Name	Intake of calcium (g. per day)			Excretion of calcium (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Excretion in urine as per cent of intake
		Bread and flour	Other foods	Total	Urine	Faeces	Total				
100	Hilde	1·84	0·62	2·46	0·16	2·20	2·36	+ 0·10	0·26	10·6	6·5
	Bernhard	1·93	0·58	2·51	0·24	2·03	2·27	+ 0·24	0·48	19·1	9·6
	Erika	1·94	0·67	2·61	0·26	1·98	2·24	+ 0·37	0·63	24·1	10·0
	Average	1·90	0·62	2·52	0·22	2·07	2·29	+ 0·23	0·45	17·9	8·7
85	Helga	0·59	0·62	1·21	0·13	0·87	1·00	+ 0·21	0·34	28·1	10·7
	Horst	0·96	0·55	1·51	0·22	1·16	1·38	+ 0·13	0·35	23·2	14·5
	Agnes	0·99	0·67	1·66	0·35	1·04	1·39	+ 0·27	0·62	37·3	21·1
	Average	0·85	0·61	1·46	0·23	1·02	1·25	+ 0·21	0·44	30·1	15·8
70	Marianne	0·47	0·62	1·09	0·23	0·56	0·79	+ 0·30	0·53	48·6	21·1
	Helmut	0·59	0·58	1·17	0·30	0·55	0·85	+ 0·32	0·62	53·0	25·6
	Irmtraud	0·61	0·67	1·28	0·10	0·85	0·95	+ 0·33	0·43	33·6	7·8
0 enriched to 100	Hannelore	0·54	0·74	1·28	0·30	0·53	0·83	+ 0·45	0·75	58·6	23·4
	Harro	0·73	0·58	1·31	0·17	0·85	1·02	+ 0·29	0·46	35·1	13·0
	Ursula	0·56	0·67	1·23	0·20	0·87	1·07	+ 0·16	0·36	29·2	16·2
0 enriched to 85	Hanni	0·36	0·62	0·98	0·15	0·80	0·95	+ 0·03	0·18	18·4	15·3
	Günther	0·57	0·58	1·15	0·24	0·54	0·78	+ 0·37	0·61	53·0	20·9
	Frieda	0·63	0·67	1·30	0·35	0·69	1·04	+ 0·26	0·61	46·9	26·9
	Average all 70 per cent flours	0·56	0·64	1·20	0·23	0·69	0·92	+ 0·28	0·51	42·5	19·2
	Average excluding 'Hanni'	0·59	0·64	1·23	0·24	0·68	0·92	+ 0·31	0·55	44·7	19·5

calculated that British children of similar ages received from their whole diet before the war. The German diets contained very little milk, and this 0·6–0·7 g. of calcium must have come largely from the cooked green vegetables and soups which formed the bulk of the non-cereal part of the children's diet (see Holman, p. 92).

The amount of calcium absorbed varied from one child to another, but the averages for the groups having bread made from flours of 100 per cent, 85 per cent and 70 per cent extractions were similar, as also were the amounts excreted in the urine and the amounts retained. The quantities of calcium added to the flour were based on the recommendations of McCance and Widdowson (1942), and it appears that these were in the correct proportions to produce similar absorptions and retentions whatever the extraction rate of the flour.

The most comprehensive series of calcium balance studies on well-nourished children are those of Macy and her co-workers (Macy, 1942). Her children were living on "foods most frequently recommended for the dietaries of children", which included 500 g. of milk a day. The average calcium intake of her two oldest children, aged 11 and 12 years, was 0·95 g. a day. These children absorbed 0·28 g. and retained 0·17 g. a day. Thus the German children had more calcium in their diets than Macy's children, and they absorbed more from their food. They excreted more in the urine, and the final retentions were similar to those of Macy's subjects, though tending to be rather higher.

A full-term baby contains about 28 g. of calcium (Widdowson and Spray, 1951), and an adult about 1200 g. (Widdowson, McCance and Spray, 1951). If it is assumed that a child has 18 years in which to lay down 1172 g. of calcium, he has to retain an average of 0·18 g. a day, which is very close to Macy's figure. Although the German children had been living on the experimental diets for some months before any metabolic studies were made, most of them were still below the standard height for their ages (Table 1). They were all growing faster than the standard rate, and it was only to be expected that, given the opportunity, they would retain more calcium than Macy's well-grown children. The chief importance of the results, however, lies in the fact that the children were able to absorb and retain calcium so well on all the diets. How well they would have fared had they not been given additional calcium in their bread is an interesting point.

There is some information about this in the results of a study made by Widdowson and Thrussell (1951) on some children in a smaller orphanage in Germany. These children were living on their German rations, with additional bread made from flour of 100 per cent extraction. Half received bread containing added calcium carbonate, so that their total calcium intakes were about 2·5 g. a day. The others had no extra calcium, and their intakes were 0·75 g. a day. After the children had been eating these diets for 4 months the calcium ingested and excreted by some of the children in each group was measured. The first group retained an average of 0·34 g. of calcium a day, which was more than the 0·17 g. that the children in the second group were able to absorb from their food. This second group had a positive balance of only 0·085 g. a day. Had the bread used in the Duisburg experiments not been fortified with calcium, the total calcium intakes of the children taking part in the metabolic studies would have been about 0·70–0·75 g. a day. The children receiving bread made from flour of 70 per cent extraction might have fared reasonably well, but those having the 100 per cent extraction flour would undoubtedly have absorbed less calcium than they did. They would have excreted less in the urine, but it seems unlikely that their retentions could ever have been as much as 0·18 g. a day. In other words, their positive balance would not have been sufficient to enable them to lay down calcium at a rate which would have allowed their bones to develop normally.

Phosphorus

The intakes of phosphorus (Table 6) varied between 1 and 2 g. a day according to the extraction rate of the flour, but the amounts of calcium carbonate which

TABLE 6

Absorption and excretion of phosphorus on the experimental diets at Duisburg

xtraction rate of flour (per cent)	Name	Intake of phosphorus (g. per day)			Excretion of phosphorus (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Excretion in urine as per cent of intake
		Bread and flour	Other foods	Total	Urine	Faeces	Total				
100	Hilde	1·56	0·40	1·96	0·60	1·30	1·90	+ 0·06	0·66	33·7	30·6
	Bernhard	1·79	0·53	2·32	0·68	1·57	2·25	+ 0·07	0·75	32·3	29·3
	Erika	1·69	0·46	2·15	0·75	1·09	1·84	+ 0·31	1·06	49·3	34·9
	Average	1·68	0·46	2·14	0·68	1·32	2·00	+ 0·14	0·82	38·3	31·8
85	Helga	0·60	0·40	1·00	0·53	0·44	0·97	+ 0·03	0·56	56·0	53·0
	Horst	1·11	0·51	1·62	0·57	0·84	1·41	+ 0·21	0·78	48·1	35·2
	Agnes	1·01	0·46	1·47	0·76	0·48	1·24	+ 0·23	0·99	67·3	51·7
	Average	0·91	0·46	1·37	0·62	0·59	1·21	+ 0·16	0·78	56·9	45·3
70	Marianne	0·43	0·40	0·83	0·44	0·27	0·71	+ 0·12	0·56	67·5	53·0
	Helmut	0·70	0·53	1·23	0·69	0·35	1·04	+ 0·19	0·88	71·5	56·1
	Irmtraud	0·65	0·46	1·11	0·43	0·41	0·84	+ 0·27	0·70	63·1	38·7
0 enriched to 100	Hannelore	0·50	0·47	0·97	0·52	0·26	0·78	+ 0·19	0·71	73·2	53·6
	Harro	0·75	0·53	1·28	0·73	0·55	1·28	± 0	0·73	57·0	57·0
	Ursula	0·52	0·46	0·98	0·45	0·42	0·87	+ 0·11	0·56	57·2	45·9
0 enriched to 85	Hanni	0·32	0·40	0·72	0·54	0·33	0·87	− 0·15	0·39	54·2	75·0
	Günther	0·61	0·53	1·14	0·71	0·33	1·04	+ 0·10	0·81	71·1	62·3
	Frieda	0·62	0·46	1·08	0·51	0·37	0·88	+ 0·20	0·71	65·7	47·2
	Average all 70 per cent flours	0·57	0·47	1·04	0·56	0·36	0·92	+ 0·12	0·67	64·4	53·8
	Average excluding 'Hanni'	0·60	0·48	1·08	0·56	0·37	0·93	+ 0·15	0·71	65·7	51·9

had been added to the flours were such that the children in all groups absorbed 0·7–0·8 g. of phosphorus a day and retained about 0·15 g. These results may be compared with those obtained by Macy for 11 and 12-year old children. Macy's children had 1·5 g. of phosphorus a day in their food; they absorbed more than any of the German children, presumably because of their lower calcium intakes, and they excreted more in the urine. Their average retention of phosphorus (0·12 g. a day), like their retention of calcium, was a little lower than that of the German children.

Table 7 shows the average ratio of Ca/P in the food, urine and faeces of the children taking part in the present study, and the results for the 'absorption' and retention have also been expressed in this way. In all three groups the Ca/P ratio of the diet was over 1; this promoted a retention ratio which was in general as high as that observed by Macy for children receiving 500 g. of milk a day.

Magnesium

The magnesium intakes (Table 8), like those of phosphorus, varied with the extraction rate of the flour. Children eating white flour had 0·3 g. magnesium

TABLE 7

Average Ca/P ratios

Extraction rate of flour (per cent)	Intake Ca/P	Ca/P in urine	Ca/P in faeces	Absorption Ca/P	Retention Ca/P
100	1·18	0·32	1·57	0·56	1·65
85	1·07	0·37	1·73	0·56	1·25
70	1·14	0·43	1·84	0·78	2·07
Macy's (1942) figures	0·63	0·13	1·56	0·26	1·40

TABLE 8

Absorption and excretion of magnesium on the experimental diets at Duisburg

Extraction rate of flour (per cent)	Name	Intake of magnesium (g. per day)			Excretion of magnesium (g. per day)			Balance (g. per day)	Absorption (g. per day)	Absorption as per cent of intake	Excretio in urine as per cer of intak
		Bread and flour	Other foods	Total	Urine	Faeces	Total				
100	Hilde	0·686	0·139	0·825	0·155	0·621	0·776	+0·049	0·204	24·7	18·8
	Bernhard	0·674	0·126	0·800	0·121	0·674	0·795	+0·005	0·126	15·8	15·1
	Erika	0·673	0·138	0·811	0·181	0·607	0·787	+0·024	0·204	25·2	22·2
	Average	0·678	0·134	0·812	0·152	0·634	0·786	+0·026	0·178	21·9	18·7
85	Helga	0·236	0·139	0·375	0·124	0·224	0·348	+0·027	0·151	40·3	33·1
	Horst	0·349	0·120	0·469	0·102	0·316	0·418	+0·051	0·153	32·6	21·8
	Agnes	0·359	0·138	0·496	0·179	0·296	0·475	+0·021	0·200	40·3	36·1
	Average	0·315	0·132	0·447	0·135	0·279	0·414	+0·033	0·168	37·6	30·2
70	Marianne	0·131	0·139	0·270	0·111	0·145	0·256	+0·014	0·125	46·3	41·1
	Helmut	0·180	0·125	0·305	0·147	0·113	0·260	+0·045	0·192	63·0	48·2
	Irmtraud	0·171	0·138	0·309	0·141	0·151	0·292	+0·017	0·158	51·1	45·6
70 enriched to 100	Hannelore	0·175	0·171	0·346	0·102	0·204	0·306	+0·040	0·142	41·0	29·5
	Harro	0·221	0·125	0·346	0·122	0·158	0·280	+0·066	0·188	54·3	35·3
	Ursula	0·151	0·138	0·289	0·094	0·184	0·278	+0·011	0·105	36·3	32·5
70 enriched to 85	Hanni	0·093	0·139	0·232	0·120	0·172	0·292	−0·060	0·060	25·8	51·7
	Günther	0·155	0·125	0·280	0·127	0·139	0·266	+0·014	0·141	50·4	45·4
	Frieda	0·193	0·138	0·331	0·075	0·218	0·293	+0·038	0·113	34·1	22·6
	Average all 70 per cent flours	0·163	0·138	0·301	0·115	0·165	0·280	+0·021	0·136	45·2	38·2
	Average excluding 'Hanni'	0·172	0·137	0·309	0·115	0·164	0·279	+0·030	0·145	46·9	37·2

a day; those having wholemeal flour 0·8 g. The amounts absorbed varied with the intakes, as did also the amounts excreted in the urine. The average retention of magnesium, like that of calcium and phosphorus, was the same in all groups. The figure was almost identical with Macy's value for the magnesium retention of 11 and 12-year old children whose diets provided them with 0·3 g. of magnesium a day.

Conclusions

The results obtained in this series of metabolic balance studies show that the children absorbed and retained fully as much nitrogen, calcium, phosphorus and magnesium from their high-bread diets as American children of similar ages have been shown to do from more conventional diets. From this point of view, therefore, the experimental diets must be considered satisfactory.

The children receiving the flour of 100 per cent extraction had the highest intakes of all four elements, but the average amount they retained was almost exactly the same in every case as the average amount retained by children having 85 per cent or 70 per cent extraction flour.

Summary

1. Metabolic studies were made on 3 children from each bread group at Duisburg and on 2 boys from each group at Vohwinkel after the children had been eating the experimental diets for 3–7 months. The intakes and excretions of nitrogen were measured at both orphanages, and those of calcium, phosphorus and magnesium at Duisburg.

2. The average nitrogen retentions were similar for all groups at both homes and amounted to about 2 g. a day.

3. The quantities of calcium carbonate which had been added to the experimental flours, together with the calcium from the rest of the diet, enabled the children in all groups at Duisburg to retain 0·2–0·3 g. calcium a day.

4. The retentions of phosphorus and magnesium were also satisfactory.

References

McCance, R.A. and Walsham, C. M. (1948). The digestibility and absorption of the calories, proteins, purines, fat and calcium in wholemeal wheaten bread. *Brit. J. Nutr.*, **2**, 26.

McCance, R. A. and Widdowson, E. M. (1942). Mineral metabolism of healthy adults on white and brown bread dietaries. *J. Physiol.*, **101**, 44.

McCance, R. A. and Widdowson, E. M. (1946). An experimental study of rationing. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 254.

Macy, I. G. (1942). *Nutrition and chemical growth in childhood.* Vol. I. Evaluation. Charles C. Thomas, Springfield, U.S.A.

Meyer, G. (1871). Ernährungsversuche mit Brod am Hund und Menschen. *Z. Biol.*, **7**, 1.

Newman, L. F., Robinson, G. W., Halnan, E. T. and Neville, H. A. D. (1912). Some experiments on the relative digestibility of white and wholemeal breads. *J. Hyg., Camb.*, **12**, 119.

O'Brien, R., Girshick, M. A. and Hunt, E. P. (1941). Body measurements of American boys and girls for garment and pattern construction. *Misc. Publ. U.S. Dep. Agric.* No. 366. Washington, D.C.

Rubner, M. (1883). Ueber den Werth der Weizenkleie für die Ernährung des Menschen. *Z. Biol.*, **19**, (N.F. 1), 45.

Rubner, M. (1917). Untersuchungen über Vollkornbrote. *Arch. Anat. Physiol., Physiol. Abt.*, p. 255.

Snyder, H. (1905). Studies of the digestibility and nutritive value of bread and of macaroni at the University of Minnesota 1903-1905. *U.S. Dept. Agr. Off. Expt. Sta. Bull.* 156.

Snyder, H. (1908). *Human foods and their nutritive value.* Macmillan and Co., New York.

Widdowson, E. M. (1947). A study of individual children's diets. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 257.

Widdowson, E. M. (1951). Mental contentment and physical growth. *Lancet*, **1**, 1316.

Widdowson, E. M., McCance, R. A. and Spray, C. M. (1951). The chemical composition of the human body. *Clin. Sci.*, **10**, 113.

Widdowson, E. M. and Spray, C. M. (1951). Chemical development in utero. *Arch. Dis. Childh.*, **26**, 205.

Widdowson, E. M. and Thrussell, L. A. (1951). Studies of undernutrition, Wuppertal 1946–9. 26. The absorption and excretion of nitrogen, calcium, magnesium and phosphorus. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 275.

APPENDIX C: BIOCHEMICAL INVESTIGATIONS INTO THE B-VITAMIN METABOLISM OF CHILDREN HAVING THE EXPERIMENTAL DIETS

by W. I. M. Holman

Introduction

In the past fifteen years much attention has been paid to the question of whether white wheaten bread can provide adequate amounts of B-vitamins for full nutritional needs. Although the position with regard to some of the B-factors is still far from clear, available evidence suggests that a diet of white bread alone might not contain sufficient aneurin in relation to its energy value to satisfy human requirements (Holman, 1946). This conclusion was reached by a consideration of the average amounts of aneurin in flours of 60 to 70 per cent extraction, the losses of aneurin in bread-making, and the estimated aneurin requirements of adults. Direct studies of the effects on human subjects of diets consisting largely of white bread had not been made.

Since it seemed possible that the children eating unenriched white bread in the present investigations might not receive adequate supplies of B-vitamins, at any rate of aneurin, and might later show signs of deficiency, it was considered desirable that their B-vitamin status should be studied by biochemical methods during the experiments. A warning could then be given of approaching deficiency, before the clinical signs became obvious, and before the health of the children was endangered. There appeared to be little risk that the children eating the other types of bread would suffer from deficiencies of this nature, but studies of their B-vitamin metabolism would be of value as a means of establishing whether any differences observed in the clinical status of the children were due to the vitamins, or to other dietary factors. The results would also help to show whether the vitamins in the various sorts of bread were equally well utilized by the children. Since only aneurin, riboflavin and nicotinic acid were added in the preparation of the enriched white flours, the investigations were confined to these three vitamins.

Methods

EXPERIMENTAL PROCEDURE

Of the various biochemical methods which have been proposed for assessing the state of nutrition with respect to B-vitamins, probably the most satisfactory are those based on the determination of the 24-hour urinary excretion of the vitamins or their metabolites after the subject has received a known intake (Mason and Williams, 1942; Benson, Witzberger and Slobody, 1943). No generally accepted method is available, however, since much has yet to be learned about the metabolism of the B-vitamins, especially of riboflavin and nicotinic acid (Nutrition Reviews, 1947a, b; Williams, Eakin, Beerstecher and Shive, 1950). Moreover, it is still not clear whether B-vitamins synthesized by intestinal micro-organisms are normally absorbed and utilized. The levels of the vitamins in whole blood do not appear to be a useful guide to the state of nutrition and cannot be used to indicate the existence of sub-clinical deficiency (Klein, Perlzweig and Handler, 1942; Benson *et al.*, 1943). Procedures involving the administration of test doses of the vitamins could not be used in the present experiments because they might have had some influence on the outcome of the

main experiment. In view of these considerations it seemed desirable to make a thorough examination of the children's intakes and of their urinary and faecal excretions of the vitamins while they were living on the experimental diets.

Before the children at the orphanages came under observation, their B-vitamin intake must have been high, judging by the nature of the foods which were being eaten. In order to reduce the intake, unenriched white bread was provided throughout the preliminary period, instead of the dark brown German bread. Later in this period the other foods in the diet were analysed for aneurin, and those which gave the highest values were either used in smaller quantities, or, if possible, were omitted altogether. The intake of B-vitamins varied during the preliminary period, partly as a result of these modifications in the diet, and also because strict control over the children's food was not easy to ensure until the experimental routine had become established. To give an indication of the state of nutrition at this time, 24-hour specimens of urine were collected from some of the children and analysed for B-vitamins.

After the commencement of the experimental period at each orphanage, the 24-hour urinary excretions of the vitamins by children in each bread group were determined from time to time as a general guide to their vitamin status. In addition, metabolic studies were made in order to provide detailed information about their intakes and their urinary and faecal excretions. These studies were made when the children had received the experimental breads for periods of from 3 to 7 months. Ample time was thus allowed for the effects of the different types of bread on the B-vitamin metabolism of the children to become established. Each child was studied over a period of 7 days, the intakes and excretions of nitrogen and minerals being determined at the same time. Details of the children chosen for the metabolic studies and the experimental methods adopted have already been given on pp. 21, 83.

PRESERVATION OF SAMPLES FOR ANALYSIS

Owing to the labile nature of some of the B-vitamins, and to the possibility that they might be synthesized by micro-organisms, all analyses were made as soon as possible after the samples had been collected. Since some delay was unavoidable, however, particularly in the metabolic studies, the following precautions were also taken. Soup, vegetables, fruit and drinks were preserved with acetic acid and toluene, and faeces with hydrochloric acid and toluene. The samples were kept in a refrigerator until they were analysed. Bread was dried *in vacuo* at a temperature of 35° C. as soon as it was received and was stored in the dry state. Samples of urine for aneurin and N-methylnicotinamide determinations were preserved with acetic acid and toluene and were kept in a refrigerator. Samples for the determination of riboflavin were preserved with toluene alone and were stored in the dark at room temperature (see Slater and Morell, 1946b).

METHODS OF ANALYSIS

The vitamin determinations were performed by German technicians, each of whom was trained to carry out one of the methods. All the determinations of a particular vitamin were made by the same individual under strict supervision.

The fluorimeter used in the determination of aneurin, riboflavin and N-methylnicotinamide was designed to be independent of mains voltage fluctuations, which at that time were frequent in Wuppertal. The principles of its construction

were as follows. A 125-watt Hg vapour lamp was used as the light source, a 'Variac' variable voltage transformer being included in the circuit to enable the light intensity to be altered. The lamp was mounted in a housing provided with two apertures through which two similar horizontal beams of light were emitted from the centre of the arc. One beam was directed on to a selenium photocell specially made for use with ultraviolet light by Evans Electroselenium, Limited. A micro-ammeter attached to the photocell registered any change which occurred in the light intensity. By appropriate adjustment of the transformer knob the reading of the micro-ammeter could be set and kept constant at any particular value. Even a rapid fluctuation could be controlled in this way. In order to prevent fatigue of the photocell, a shutter was placed between the lamp and photocell and was opened only while fluorescence measurements were being made.

The second beam of light was focused by a quartz lens and directed from side to side through a rectangular glass cell containing the test solution. The design of this part of the instrument was based on systems commonly employed in fluorimetry. The fluorescent light, emerging from the glass cell in a direction at right angles to the activating beam, passed through a diaphragm on to a second photocell, which was connected to a mirror galvanometer. The galvanometer scale was placed immediately below the micro-ammeter so that both scales could be viewed in rapid succession. Slots for primary filters were provided in front of the test solution and the control photocell, and those for secondary filters in front of the photocell receiving the fluorescent light. The lamp was switched on 30 min. before the instrument was used in order to obtain a high and constant proportion of light of low wavelengths. All readings of the galvanometer were taken at a standard reading of the micro-ammeter and were thus strictly comparable.

Aneurin

A modification of the method of Greenberg and Rinehart (1945) was used. Each sample of food or faeces was extracted for 20 min. in a boiling water bath with a 0·5 per cent solution of HPO_3, and the extract was adjusted to pH 4·5 and incubated with takadiastase to convert co-carboxylase to the free vitamin. Urine was adjusted to pH 4·5, but was not treated with takadiastase. Ten ml. of the extract or diluted urine, containing from about 0·5 to 1 μg. of aneurin, were passed through a column of 'Decalso'. The amount of 'Decalso' used was 2 g. and the diameter of the column 1 cm. The adsorbent was of the highest quality obtainable. The adsorbed aneurin was eluted with an acidified 25 per cent solution of KC1 in the manner described by Greenberg and Rinehart, and the aneurin in the eluate was oxidized to thiochrome by alkaline ferricyanide in the presence of methanol. The conditions were rigidly standardized. The blank was prepared by the omission of ferricyanide. The relationship between the galvanometer reading and the amount of aneurin present was linear. An internal standard was included in each determination by adding a known amount of aneurin to a portion of the extract or diluted urine before adsorption, and carrying out a second analysis on this solution.

As Najjar and Ketron (1944) have pointed out, the thiochrome method gives results which are too low when N-methylnicotinamide is present, e.g. in the determination of aneurin in urine. However, since the error varies directly with the amount of N-methylnicotinamide present, and since N-methylnicotinamide

was determined in every sample of urine which was analysed for aneurin, it was possible to apply a correction. The correction factor used, which was determined by experiment, was as follows:

$$\begin{matrix}\text{Corrected aneurin} \\ \text{excretion} \\ \text{(mg. per 24 hr.)}\end{matrix} = \begin{matrix}\text{Uncorrected} \\ \text{value} \\ \text{(mg. per 24 hr.)}\end{matrix} + \left(\begin{matrix}0{\cdot}009 \times \text{N-methylnicotinamide} \\ \text{excretion} \\ \text{(mg. per 24 hr.)}\end{matrix}\right)$$

Riboflavin

The method of Slater and Morell (1946a) was used because of its specificity. An artificial source of ultraviolet light, instead of sunlight, was used for the destruction of riboflavin in the pyridine-butanol extracts. The tubes containing the extracts were placed in a circular rack which could be made to rotate slowly, by means of a small electric motor, around an ultraviolet lamp. In this way all extracts received the same amount of irradiation. An internal standard was included in each determination in the manner described by Slater and Morell. The relationship between the galvanometer deflexion and the amount of riboflavin present was linear.

The accuracy of the method of Slater and Morell when applied to urine has been confirmed by Ritter, Moore, Hirschberg and Rubin (1949). In order to ensure complete extraction of riboflavin from foods, preliminary tests were made using various periods of autoclaving with 0·25 N H_2SO_4 or of incubation with papain and takadiastase. It was found that, although similar results were obtained for the non-cereal part of the orphanage diet by a variety of methods, only when samples of flour were autoclaved with H_2SO_4 for at least 30 min. did the values reach levels comparable with those given under the same conditions by the dried bread made from the flour. All samples of foods and faeces were therefore extracted by method B of Slater and Morell, i.e. they were autoclaved in 0·25 N H_2SO_4 solution for 30 to 35 min. at 120° and allowed to cool slowly.

Nicotinic Acid

Owing to the limitations of knowledge when this work began in 1947, the selection of a method presented difficulties. In the case of foods, particularly cereals, it is still uncertain what derivatives other than the acid and its amide are determined by the usual methods, and whether they are utilizable by man. Chaudhuri and Kodicek (1950a, b) have suggested that the bulk of the nicotinic acid in wheat bran is present as a precursor, which is readily hydrolysable to nicotinic acid by alkalis and more slowly by acids, but which may not be utilizable. It was decided in the present experiments to determine the total acid-hydrolysable derivatives of nicotinic acid, in urine by the method of Perlzweig, Levy and Sarett (1940), and in foods and faeces by a slight modification of the same method which has been applied to various biological materials, including cereals (Dann and Handler, 1941). In the case of urine, it was shown by Perlzweig *et al.* that the method gives the total nicotinic acid derived from nicotinic acid, nicotinamide and nicotinuric acid. It does not include quinolinic acid (Holman, unpublished data).

It has been shown that the human infant (Snyderman, Ketron, Carretero and Holt, 1949) and adult (Holman and de Lange, 1950a) can convert tryptophan to nicotinic acid to an important extent, but it is not yet possible to estimate the amounts which are normally obtained in this way.

N-methylnicotinamide was determined in urine by the method of Huff and Perlzweig (1947) and N-methyl-2-pyridone-5-carboxylamide by a procedure similar to that described by Holman and de Lange (1949). Since evidence is available (Ellinger, 1947) that N-methylnicotinamide may be present in the faeces of the rat, determinations of this metabolite were also made in some of the samples of faeces by the fluorimetric method of Huff and Perlzweig (1947). The results are shown in Table 1. Since this method was worked out only for urine, an attempt was made to check the results by the colorimetric method of Sarett (1943) for total N-methyl derivatives of nicotinic acid, but no detectable colour was produced. Owing to this failure to confirm the results obtained by the fluorimetric method, no allowance for faecal N-methylnicotinamide was made in calculations of the total output of nicotinic acid and aneurin (Tables 9 and 11, pp. 110, 112). The extent to which the amounts of N-methylnicotinamide found in faeces would affect the balances of these two vitamins is indicated in Table 1.

TABLE 1

Amounts of N-methylnicotinamide in faeces as estimated by the method of Huff and Perlzweig, and their influence on the aneurin and nicotinic acid balances

Extraction rate of flour (per cent)	N-methyl-nicotinamide in faeces (mg. per day)	Aneurin balance (mg. per day)		Nicotinic acid balance† (mg. per day)	
		Uncorrected*	Corrected	Uncorrected*	Corrected
		Duisburg			
100	4·8	+ 0·81	+ 0·77	+ 14·9	+ 10·6
85	2·1	+ 0·99	+ 0·97	+ 10·2	+ 8·3
70	2·0	+ 0·85	+ 0·83	+ 9·1	+ 7·3
70 enriched to 100	0·8	+ 1·14	+ 1·13	+ 19·3	+ 18·6
70 enriched to 85	0·9	+ 0·80	+ 0·79	+ 14·2	+ 13·4
		Vohwinkel			
100	2·3	+ 0·80	+ 0·78	+ 15·6	+ 13·5
	2·7	+ 0·31	+ 0·29	+ 12·5	+ 10·1
70	1·7	+ 0·67	+ 0·65	+ 2·6	+ 1·1
	0·7	+ 0·52	+ 0·51	+ 3·8	+ 3·2
70 enriched to 100	1·0	+ 0·94	+ 0·93	+ 11·3	+ 10·4
	1·7	+ 1·02	+ 1·00	+ 10·6	+ 9·1

* Values obtained when no allowance is made for faecal N-methylnicotinamide (see Tables 9 and 11).

† Not including urinary excretion of N-methyl-2-pyridone-5-carboxylamide.

Composition of the Experimental Flours and of the Breads made from them

The unenriched white flour received by the children during the preliminary period at Duisburg and in the first half of the preliminary period at Vohwinkel was not the same as that used subsequently at the two orphanages. Samples taken from different sacks were analysed and found to range in aneurin content from 1·6 to 2·3 μg. per g. Independent analyses made on the same samples by a thiochrome method at the Cereals Research Station, St. Albans, gave values of from 1·4 to 2·3 μg. per g. This flour was richer in aneurin, and more variable in composition, than the 70 per cent extraction flour used in the experimental periods, but since it was used only in the preliminary periods its exact nature was not of great importance in the experiments. In one sample the concentrations of aneurin, riboflavin and nicotinic acid were determined by the methods described in the previous section and were found to be 1·7, 0·40 and 16 μg. per g., respectively.

The five flours used in the first 6 months of the experimental period at Duisburg were a consignment specially prepared from the same grist. Details about the methods of preparation have already been given (see p. 13). In the second 6 months at Duisburg and throughout the experimental period at Vohwinkel, a second consignment of each of these flours was used, made from another grist, but prepared by methods as similar to those used for the preparation of the first batch as possible. Some of the second consignment of unenriched white flour was used in the second half of the preliminary period at Vohwinkel. Before each of these flours was used, it was carefully sampled (see p. 13 and footnote, Table 2) and analysed for B-vitamins. Confirmatory analyses for aneurin and riboflavin were made on some of the samples at the Cereals Research Station, St. Albans. The results are given in Table 2. They show that the concentrations of B-vitamins in the second consignment were not identical with those in the first, but that the differences were not great. In both consignments higher values were found for riboflavin, especially by the chemical method, in the wholemeal flour than in its enriched analogue. In other respects, however, the concentrations of B-vitamins in the enriched flours were similar to those in the flours they were intended to imitate. In view of the difficulties of B-vitamin analysis and the magnitude of the deviations between individual results observed in other collaborative studies of B-vitamin methods (see Vitamin B_1 Subcommittee of the Accessory Food Factors Committee of the Medical Research Council, 1943; Andrews, 1943), the agreement between the results obtained at Wuppertal and those obtained by different methods at St. Albans is reasonably good. Independent analyses for nicotinic acid were not made on these samples, but microbiological assays, made before the flours were despatched, gave lower results in almost all cases (10 μg. per g. for the unenriched white flour, 26 μg. per g. for the 85 per cent and corresponding enriched flour, and 48 μg. per g. for the wholemeal flour and its enriched counterpart).

When compared with other values reported in the literature, the concentrations of aneurin found in the flours of 100, 85 and 70 per cent extraction appear to be within the normal ranges for flours of these types. In the case of the 70 per cent extraction flour, however, the values tend to be at the upper limit of the range (taken as 0·8 to 1·5 μg. per g.; see Holman, 1946, for references), possibly due to the fact that the flour was prepared in a mill used normally at that time for the production of 85 per cent extraction flour; traces of scutellum and bran

may have been introduced, thus tending to raise the concentration of aneurin slightly. The mixture of wheats used for the preparation of the flour was not unusual (see p. 13). The results for riboflavin in the 70 per cent extraction

TABLE 2

Concentrations of B-vitamins in the experimental flours and in the dried bread made from the flours

Sample	Dry matter (g. per 100 g.)	Aneurin (μg. per g.)	Riboflavin (μg. per g.)	Nicotinic acid derivatives (μg. per g.)
Flour				
1st consignment*				
100 per cent extraction ..	88·2	4·25 (4·1‡)	1·50 (1·24‖)	45
85 per cent extraction ..	87·8	3·6 (3·5)	0·81 (0·93)	31
70 per cent extraction ..	87·6	1·4 (1·1)	0·41 (0·47)	19
70 per cent extraction enriched to 100 per cent levels	87·6	4·45 (3·8)	1·02 (1·06)	55
70 per cent extraction enriched to 85 per cent levels	86·9	3·2 (2·9)	0·71 (0·83)	33
2nd consignment†				
100 per cent extraction ..	—	4·0	1·53	56
85 per cent extraction ..	—	3·1	0·80	27
70 per cent extraction ..	—	1·6	0·50	19
70 per cent extraction enriched to 100 per cent levels	—	3·8	0·94	56
70 per cent extraction enriched to 85 per cent levels	—	3·6	0·70	34
Dried bread				
1st consignment				
100 per cent extraction ..	90·0	4·1 (2·8‡)	1·64 (1·83‖, 2·9§)	47
85 per cent extraction ..	89·8	3·2 (3·2)	0·80 (1·50, 2·48)	33
70 per cent extraction ..	91·2	1·75 (1·7)	0·35 (0·68, 1·6)	19
70 per cent extraction enriched to 100 per cent levels	89·6	3·65 (3·0)	1·10 (1·26, 2·25)	54
70 per cent extraction enriched to 85 per cent levels	89·2	2·6 (2·6)	0·81 (0·83, 1·35)	33

* Composite samples prepared by taking 5 kg. flour from each of six sacks and mixing well.
† Composite samples prepared by taking 2 kg. flour from each of ten sacks.
‡ Independent analyses made by a thiochrome method at the Cereals Research Station, St. Albans, are shown in the brackets in this column.
‖ Independent analyses made by a microbiological method at the Cereals Research Station, are shown in the brackets in this column.
§ Rat growth assays made at the Lister Institute, London, are shown as the second figure in the brackets in this column.

flour were slightly higher than those reported by other investigators (0·35–0·40 μg. per g.; see Kodicek, 1946).

Since the composition of the breads was of even greater importance to the experiments than that of the flours from which they were made, the methods used by the German bakers at Duisburg and Vohwinkel were scrutinized. Their usual recipe was stated to be: flour 1·75 kg.; water *ca.* 1 litre; salt 30 g.; yeast 25–30 g. (1·4–1·7 per cent of the weight of flour). The baking time was 50 min. for the wholemeal bread and 40 min. for the other breads. Calcium acetate was used on several occasions in the first few weeks of the Duisburg experiment to prevent 'rope' when the weather was hot, but the necessity for using it was subsequently avoided by baking the breads freshly each day. The yeast used by both bakers was prepared under standard conditions by a process similar to that used in England. The manufacturer provided the following data on its

TABLE 3

Concentrations of B-vitamins in the yeast and in the fresh bread

Sample	Dry matter (g. per 100 g.)	Aneurin (μg. per g.)	Riboflavin (μg. per g.)	Nicotinic acid derivatives (μg. per g.)
Yeast, fresh	25–30	8·0	9·7	107
Bread, fresh*				
100 per cent extraction ..	55·4	2·5 (2·7†)	1·01 (1·02†)	29 (29†)
85 per cent extraction ..	56·6	2·0 (2·3)	0·50 (0·60)	21 (21)
70 per cent extraction ..	57·6	1·1 (1·0)	0·22 (0·36)	12 (13)
70 per cent extraction enriched to 100 per cent levels	58·1	2·4 (3·0)	0·71 (0·76)	35 (37)
70 per cent extraction enriched to 85 per cent levels	58·4	1·7 (2·2)	0·53 (0·56)	22 (23)

* Made from the first consignment of experimental flours.
† Theoretical values, calculated from the B-vitamin contents of the flour and the yeast, assuming that no synthesis or destruction of B-vitamins occurred during breadmaking, are shown in the brackets.

composition: water 70–75 per cent; protein 10–15 per cent; ash 1–3 per cent; P_2O_5 0·8–1·5 per cent. The concentrations of B-vitamins in the yeast were determined and are shown in Table 3.

As a check on the uniformity of the experimental flours and the bakers' methods, samples of the five different sorts of breads which were eaten by the children were collected from time to time during the experiments and analysed for B-vitamins. The concentrations in each particular sort of bread varied within reasonably narrow limits, except on one occasion at Duisburg when it was suspected that the baker had substituted another flour for the unenriched white flour (see p. 102).

The composite samples of each of the flours in the first consignment (see Table 2 for method of sampling and B-vitamin content) were baked into bread by the baker at Duisburg by a technique as similar as possible to that used in

baking for the orphanage. Five loaves of each sort of bread were dried *in vacuo* in an industrial evaporator at a temperature not exceeding 35° C. The dried breads were ground and analysed for B-vitamins. Independent analyses were made on the same samples at the Cereals Research Station, and, in the case of riboflavin, biological assays were made by a rat growth method by Miss A. M. Copping of the Lister Institute. The results are shown in Table 2 (p. 98). Except in the case of wholemeal bread, the results for aneurin at the two laboratories were in reasonably good agreement. The figure of 1·75 μg. per g. for aneurin in the dried bread made from flour of 70 per cent extraction is slightly higher than the values found for samples of the orphanage bread analysed during the experiments (range 1·2–1·6 μg. per g.). The chemical and microbiological results for riboflavin were in most cases concordant, but considerably higher values were indicated by the rat growth method than by the other two methods. Although much work has been done on riboflavin in cereals, variations of this kind between the results given by different methods of assay have still not been adequately explained (cf. Copping, 1946).

The concentrations of the vitamins in the fresh breads were calculated from the values found for the dried breads by the methods described in the previous section (see Table 2) and the moisture contents of the fresh and dried breads. The results are given in Table 3. The values shown in parenthesis in Table 3 were obtained by calculation from the concentrations of B-vitamins in the flours (Table 2) and the yeast (Table 3), on the basis of the bakers' usual recipe. A comparison between the results given by these two methods of calculation shows little evidence of an appreciable loss of riboflavin or nicotinic acid during baking. In the case of aneurin, the results indicate a loss in the breads containing synthetic aneurin, and to some extent in those made from flour of 85 or 100 per cent extraction, but not in the unenriched white bread. A loss of aneurin in bread-making has been reported by many investigators (see Holman, 1946).

B-vitamin Status of the Children during the Preliminary Period

The daily urinary excretions of B-vitamins by 6 Duisburg children aged from 7 to 14 years were determined at various stages of the preliminary period. The results are summarized in Table 4. The levels of urinary excretion at Vohwinkel were not determined until the end of the period. The results obtained at that time on 6 children, aged 13–14 years, are also included in the table. It can be seen that throughout this period the levels of urinary excretion at Duisburg were within normal limits (cf. Benson *et al.*, 1943; Williams, Mason, Cusick and Wilder, 1943; Morell and Slater, 1946; Huff and Perlzweig, 1947). There was some tendency, at least in the case of aneurin, for the urinary excretion to decrease towards the end of the preliminary period, but no striking changes occurred. At Vohwinkel the levels of excretion of aneurin and N-methylnicotinamide at the end of the preliminary period were of the same order as those observed at Duisburg, but smaller amounts of riboflavin were excreted than at Duisburg.

From these results, it seems likely that in most cases the children were receiving more than enough B-vitamins during the preliminary period to meet their needs. The supplies of riboflavin at Vohwinkel must have been somewhat restricted, but the levels of urinary excretion of the vitamin at the end of the period were not so low as to suggest any possibility of deficiency (for references see p. 107).

TABLE 4

*Urinary excretion of B-vitamins at various stages of the experiments**

ıge of experiment	Extraction rate of flour (per cent)	Urinary excretion (mg. per day)					
		Aneurin		Riboflavin		N-methylnicotin-amide	
		Range	Average	Range	Average	Range	Average
		Duisburg					
liminary period							
day after com-nencement ..	70	0·23–0·50	0·40	0·31–0·80	0·50	3·7–13·4	7·1
days	70	0·29–0·49	0·39	0·46–0·79	0·61	5·4–11·8	8·0
days	70	0·29–0·59	0·38	0·32–0·54	0·39	4·6–14·0	7·8
days	70	0·16–0·37	0·27	0·44–0·73	0·62	4·3–11·9	6·5
ɪerimental period							
year after com-ıencement ..	70	0·13–0·30	0·20	0·02–0·36	0·11	3·3–11·4	7·4
		Vohwinkel					
liminary period							
ıst day	70	0·19–0·49	0·30	0·06–0·43	0·27	3·2– 7·1	5·3
ɪerimental period							
nonths after com-ıencement ..	100	0·18–0·27	0·22	0·08–0·12	0·10	4·7–10·0	7·4
	70	0·08–0·12	0·10	0·02–0·06	0·04	4·1– 4·6	4·4
	70 enriched to 100	0·21–0·25	0·23	0·04–0·25	0·14	4·9– 6·7	5·8
months after ommencement ..	100	0·26–0·30	0·28	0·10–0·10	0·10	3·2– 6·3	4·8
	70	0·10–0·12	0·11	0·06–0·08	0·07	3·6– 3·9	3·8
	70 enriched to 100	0·31–0·32	0·32	0·22–0·45	0·34	8·7– 9·2	9·0
year after com-nencement ..	100			0·04–0·34	0·13		
	70			0·07–0·18	0·14		

* Twenty-four-hour specimens of urine were analysed. The relatively wide variation in results between ividual children is due, in part, to day-to-day fluctuations in the quantities of food eaten. A much her degree of uniformity was observed in the metabolic studies, where the period of observation was ays.

B-vitamin Intakes and Excretions during the Year of the Experiments and Interpretation and Discussion of the Findings

The results of the metabolic studies are presented in Tables 6–11. The average daily intakes and the urinary and faecal excretions at Duisburg are given in Tables 6–8 and the total output is compared with the intake in Table 9. The intakes and excretions at Vohwinkel are shown in Table 10, and the comparison of output with intake in Table 11. In order to permit the intakes and excretions to be interpreted more readily in the light of other investigations, most of which

have been made on adults, the intakes have also been calculated in relation to the energy value of the diets, and the urinary excretions as percentages of the intake. Moreover, since there is evidence that fat has a sparing action on the requirement for aneurin (e.g. see Caro and Rindi, 1950), the intakes of this vitamin have been expressed in terms of the non-fat calories of the diet as well as in terms of the total calories. These values are also included in Tables 6, 7, 8 and 10. The apparent absorption of the vitamins from the gastro-intestinal tract has been calculated by the formula:

$\frac{(\text{intake} - \text{faecal excretion}) \times 100}{(\text{intake})}$ but in view of the possibility that the vitamins may have been synthesized or destroyed in the intestinal tract, the values should be interpreted with care.

One boy at Duisburg who was eating unenriched white bread (Helmut, Tables 7 and 8) had comparatively high intakes of riboflavin and nicotinic acid, due to the fact that the unenriched white bread contained unexpectedly large amounts of these two vitamins during that particular week. Comment was also aroused about this time by the colour of the bread and too little calcium was found in it. The results for Helmut have therefore been excluded from the averages. It was suspected that the baker had substituted another white flour for some of the unenriched 70 per cent flour. The values returned to normal immediately after he had been questioned about the matter, and were normal at all other stages of the experiment. That this alteration in the bread over a period of several days had no lasting effect on the status of the children eating it is indicated by the results for Irmtraud, which were obtained at a later date (see Tables 7 and 8). They are very similar to those for the first child studied (Marianne). One of the children receiving white flour enriched to 85 per cent levels (Hanni) ate less bread than usual during the week of the metabolic studies (see p. 82). The results in this case have not been excluded from the averages. They are best interpreted in relation to the energy value of the food.

At the end of the Duisburg experiment the levels of urinary excretion of B-vitamins were determined in a group of 12 children aged 8–14 years who had eaten unenriched white bread continuously for one year. The results are given in Table 4 (p. 101). At Vohwinkel the levels of urinary excretion were determined during the first few months of the experimental period before the metabolic studies were made (see Table 4); 2 children aged 13–14 years were selected from each group for the experiments. In the case of riboflavin at Vohwinkel, the urinary excretions were also determined at the end of the experimental period in 6 children who had eaten unenriched white bread and in 6 who had eaten wholemeal bread. The ages of the children were from 11 to 15 years. The results are included in Table 4.

ANEURIN

Intake

In both experiments quite a large proportion of the total intake was supplied by foods other than bread and flour (Tables 6 and 10). The proportion was higher at Vohwinkel than at Duisburg, since smaller amounts of bread were eaten at Vohwinkel. In spite of the exclusion of many foods of high aneurin content, the non-cereal portion of the diets was comparatively rich in the vitamin in relation to its energy value. In Table 5 the average daily intakes of foods other than bread and flour by the children during the balance periods at

TABLE 5

*Average daily intakes of foods other than bread and flour during the metabolic studies**

Foodstuff	Amount of food eaten† (g. per day)	
	Duisburg	*Vohwinkel*
Milk and milk products:		
Skimmed milk	75	80
Cheese	5	1·5
Meat and meat products:		
Meat	2	0
Sausage	7	0
Fish	5	16
Eggs	2	0
Margarine	1·5	106
Sugar	0	42
Jam	65	50
Chocolate	3	2
Vegetables:		
Beans	0	186
Potatoes	127	224
Other vegetables	178	93
Soup (some containing vegetables)‡	848	472
Sauces	15	64
Fruits	26	179
Beverages:		
'Ersatz' coffee, without milk	1,180	261
Cocoa, without milk	48	0
Herb tea, without milk	0	614
Diluted orange juice	250	257

* Averages for the children aged 11–15 years who took part in the metabolic studies during the weeks in which these studies were made.

† As served.

‡ Some of the soup supplied to all children contained semolina as a thickening agent. Although strictly of cereal origin, the B-vitamins in the semolina have been included in 'other foods' and the term 'bread and flour' reserved for the experimental flours and the breads made from them.

the two orphanages are shown. Individual analyses for aneurin were not made on each of these foods, but it seems probable from the table that vegetables must have contributed most of the aneurin, for none of the cooking water was ever thrown away. It is interesting to observe that, under the conditions of food scarcity at this time in Germany, the types of food available to the orphanages and the methods of cooking were such as would tend to favour a good dietary intake of aneurin.

TABLE 6

Duisburg: Average daily intake and excretion of aneurin

Extraction rate of flour (per cent)	Name	Intake of aneurin (mg. per day)			Intake of aneurin (mg. per 1,000 Calories)		Urinary excretion of aneurin		Faecal excretion (mg. per day) (F)	$\left(\frac{I-F}{I}\right) \times$
		Bread and flour	Other foods	Total (I)	Total Cals.	Non-fat Cals.	mg. per day	per cent of intake		
100	Hilde	1·65	0·52	2·17	0·95	1·04	0·61	28	0·58	73
	Bernhard	1·78	0·50	2·28	0·93	1·01	0·50	22	0·65	72
	Erika	1·48	0·77	2·25	0·87	0·95	0·73	32	0·71	68
	Average	1·64	0·60	2·24	0·92	1·00	0·61	27	0·65	71
85	Helga	1·01	0·52	1·53	0·95	1·06	0·39	26	0·65	58
	Horst	1·34	0·47	1·81	0·77	0·83	0·42	23	0·62	66
	Agnes	1·45	0·77	2·22	0·87	0·95	0·35	16	0·88	60
	Average	1·27	0·59	1·86	0·86	0·95	0·39	21	0·72	61
70	Marianne	0·45	0·52	0·97	0·51	0·55	0·17	18	0·38	61
	Helmut	0·86	0·50	1·36	0·53	0·56	0·18	13	0·22	84
	Irmtraud	0·78	0·77	1·55	0·61	0·65	0·17	11	0·53	66
	Average, excluding 'Helmut'	0·62	0·65	1·27	0·56	0·60	0·17	13	0·46	64
70 enriched to 100	Hannelore	1·37	0·59	1·96	0·90	0·98	0·64	33	0·55	72
	Hans	1·58	0·50	2·08	0·75	0·80	0·87	42	1·14	45
	Ursula	1·24	0·77	2·01	0·88	0·95	0·49	24	0·38	81
	Average	1·40	0·62	2·02	0·84	0·91	0·67	33	0·69	66
70 enriched to 85	Hanni	0·84	0·52	1·36	0·90	0·99	0·35	26	0·70	49
	Günther	1·49	0·50	1·99	0·80	0·85	0·32	16	0·57	71
	Frieda	1·36	0·77	2·13	0·76	0·86	0·45	21	0·88	59
	Average	1·23	0·60	1·83	0·82	0·90	0·37	20	0·72	61

Estimates of the human requirements of aneurin have been made by a number of investigators. Williams, Mason, Smith and Wilder (1942) concluded from studies of the urinary excretion of the vitamin by adults that the minimum requirement is in the region of 0·4 mg. per 1,000 total Calories. On the basis of further experiments Williams, Mason and Wilder (1943) decided that the requirement is not less than 0·45 mg. per 1,000 total Calories and may be greater in the case of diets poor in fat. In their experimental diets fat supplied 33 per

cent of the total calories. Their results, therefore, suggest that the requirement is 0·66 mg. per 1,000 Calories derived from carbohydrate and protein. When expressed in this form it is independent of the amount of fat in the diet. Benson *et al.* (1943) concluded that the tissues of children are saturated with aneurin when the intake exceeds 0·4 mg. per 1,000 total Calories. An allowance of 0·5 mg. per 1,000 total Calories was recommended by the U.S. National Research Council (1948) for children aged from 13 to 15 years. Keys, Henschel, Mickelsen and Brozek (1943) observed no benefit when the intake of normal young men exceeded 0·23 mg. of aneurin per 1,000 total Calories, but their experiments were of relatively short duration (10–12 weeks). Recent investigations on young women have indicated a minimum requirement of 0·25–0·30 mg. per 1,000 total Calories (see Nutrition Reviews, 1950).

In the Duisburg experiment the intake of aneurin by the children eating the higher extraction and fortified breads was considerably greater than the highest estimated level of requirement, and the intake of the children on the unenriched white flour was approximately equal to the requirement. At Vohwinkel the amount of fat in the diets was higher than at Duisburg, and the total intakes are best compared in relation to the non-fat calories. The values indicate that the supply of aneurin at Vohwinkel was more than adequate for children eating wholemeal or enriched bread, and was just adequate for those eating unenriched white bread.

Urinary Excretion

The results of Williams, Mason and Wilder (1943) indicate that adults, who had been maintained for 8 months on a diet providing just enough aneurin to meet their minimal requirements, were excreting an average of 0·12 mg. per day in the urine. Benson *et al.* (1943) concluded that the tissues of children are saturated with aneurin if the urinary excretion is 0·15 mg. or more per day, and if 20 per cent or more of the intake is excreted in the urine. Keys, Henschel, Taylor, Mickelsen and Brozek (1945), on the basis of experiments on the restriction of the B-vitamin intakes of adults, suggested that a urinary excretion of more than 0·02 mg. of aneurin per day may indicate safety from symptoms of deficiency, but may not necessarily afford a reasonable margin in reserve.

In the light of these observations it appears that the Duisburg children eating the higher extraction and enriched flours received a supply of aneurin which was more than adequate, and that the children on unenriched white bread received enough to satisfy their requirements, but that there was less to spare. The supplies of aneurin at Vohwinkel were also adequate but, as at Duisburg, there was less to spare in the case of children on unenriched white bread. The quantities excreted by children eating unenriched white bread at Duisburg did not change significantly during the last 6 months of the experiment (cf. Tables 4 and 6).

Since the conclusions based on the urinary excretion of aneurin in every case confirm those reached from a study of the intakes, it seems reasonably certain that none of the children's diets was deficient in this vitamin.

Faecal Excretion

The faecal excretion in the Duisburg experiment tended to be lowest in children eating unenriched white bread. In all the children studied the total output was less than the intake, a finding similar to that reported for an adult

by Alexander and Landwehr (1946). The results afford no evidence as to whether aneurin was synthesized by micro-organisms in the intestinal tract or not. The apparent absorption of aneurin from the gastro-intestinal tract (see p. 102) was much the same for all the various groups of children.

The faecal excretion at Vohwinkel tended to be lower than in the corresponding groups at Duisburg, and the total output in no case exceeded the intake.

TABLE 7

Duisburg: Average daily intake and excretion of riboflavin

Extraction rate of flour (per cent)	Name	Intake of riboflavin (mg. per day)			Intake of riboflavin (mg. per 1,000 total Calories)	Urinary excretion of riboflavin		Faecal excretion (mg. per day) (F)	$\left(\frac{I-F}{I}\right) \times$
		Bread and flour	Other foods	Total (I)		mg. per day	per cent of intake		
100	Hilde	0·86	0·30	1·16	0·51	0·24	21	2·30	— 98
	Bernhard	1·02	0·42	1·44	0·58	0·14	10	2·95	— 10[illegible]
	Erika	0·91	0·44	1·35	0·52	0·32	24	2·48	— 84
	Average	0·93	0·39	1·32	0·54	0·23	17	2·58	— 95
85	Helga	0·31	0·30	0·61	0·38	0·26	43	1·14	— 87
	Horst	0·71	0·40	1·11	0·47	0·26	23	1·63	— 47
	Agnes	0·55	0·44	0·99	0·39	0·24	24	1·48	— 5[illegible]
	Average	0·52	0·38	0·90	0·41	0·25	28	1·42	— 58
70	Marianne	0·15	0·30	0·45	0·23	0·11	24	1·24	— 176
	Helmut	0·58	0·42	1·00	0·39	0·20	20	1·41	— 41
	Irmtraud	0·31	0·44	0·75	0·29	0·10	13	1·74	— 132
	Average, excluding 'Helmut'	0·23	0·37	0·60	0·26	0·11	18	1·49	— 148
70 enriched to 100	Hannelore	0·56	0·35	0·91	0·42	0·28	31	1·12	— 23
	Hans	0·71	0·42	1·13	0·41	0·33	29	1·76	— 56
	Ursula	0·39	0·44	0·83	0·36	0·23	28	1·53	— 84
	Average	0·55	0·40	0·95	0·40	0·28	30	1·47	— 55
70 enriched to 85	Hanni	0·31	0·30	0·61	0·40	0·19	31	1·02	— 67
	Günther	0·52	0·42	0·94	0·38	0·25	27	1·42	— 51
	Frieda	0·56	0·44	1·00	0·38	0·40	40	1·24	— 24
	Average	0·46	0·39	0·85	0·39	0·28	33	1·23	— 45

RIBOFLAVIN

Intake

The proportion of the total intake of this vitamin supplied by foods other than bread and flour was high (Tables 7 and 10); the differences between the total intakes of the various groups of children were therefore relatively small, particularly at Vohwinkel.

Comparatively little information is available on the human requirements of riboflavin. Williams, Mason, Cusick and Wilder (1943) and Davis, Oldham and Roberts (1946) concluded from their experiments on adults that the requirement is in the region of 0·5 mg. per 1,000 total Calories. The U.S. National Research Council (1948) recommended allowances of from 0·63 to 0·77 mg. per 1,000 total Calories for children aged 13–15 years. When compared with these estimates the riboflavin intakes of all children at both orphanages were rather low.

Urinary Excretion

The results of Williams, Mason, Cusick and Wilder (1943) indicate that adults, who had received for 8 months a diet providing just enough riboflavin to satisfy their requirements, were excreting from 0·10 to 0·14 mg. per day in the urine. It was suggested by Keys *et al.* (1945) that, for adults, a consistent urinary excretion of 0·1 mg. per day should indicate that the intake has been adequate. Davis *et al.* (1946) found that the average urinary excretions of riboflavin by women receiving increasing intakes of 0·28, 0·49 and 0·66 mg. per 1,000 total Calories were 0·11, 0·15 and 0·26 mg. per day, respectively.

When compared with these findings the levels of urinary excretion of riboflavin during the metabolic studies at Duisburg, although rather low in relation to the urinary excretions during the preliminary period and to the values given by Morell and Slater (1946) for normal urines (0·38–0·92 mg. per day), do not appear to be so low, even among children eating unenriched white bread, as to indicate deficiency of riboflavin. At Vohwinkel the levels of excretion during the metabolic studies were lower than at Duisburg, but in no case fell to zero. It appears that, at least at this stage of the Vohwinkel experiment, the riboflavin supplies of some of the children must have been marginal. At the end of the Vohwinkel experiment the levels of excretion, as judged by the results obtained over one 24-hour period, were somewhat higher than during the first 4 months. In the case of the Duisburg children eating unenriched white bread, the levels of excretion after one year on the diet were much the same as those observed in the first 6 months (compare Tables 4 and 7).

Faecal Excretion

Several investigators have studied the faecal excretion of this vitamin by adults. Davis *et al.* (1946) found that the average faecal excretion by women on diets containing 0·59–1·23 mg. of riboflavin per day (0·28–0·66 mg. per 1,000 total Calories) was 0·50–0·58 mg. per day and was independent of the intake. Denko, Grundy, Wheeler, Henderson and Berryman (1946) observed that the faecal excretion of riboflavin by adults on natural diets remained at approximately 1 mg. per day when the intake was varied from 0·32 to 1·84 mg. per day. The faecal excretion was thus about three times the intake when the latter was 0·32 mg. per day. Hathaway and Lobb (1946) reported that the faecal excretion of riboflavin by three women was much greater on a natural diet

providing 1·33 mg. per day (0·59 mg. per 1,000 total Calories) than on a synthetic diet supplying 1·09 mg. per day (0·48 mg. per 1,000 total Calories), and therefore varied with the type of food eaten. In two of the three subjects receiving the natural diet the total output of riboflavin exceeded the intake, and in one subject the faecal output alone nearly equalled the intake.

In the Duisburg experiment the levels of faecal excretion of riboflavin were high, especially for the children eating wholemeal bread. In all the children studied the faecal output of riboflavin exceeded the intake. Even when the higher intakes calculated from the rat-growth results for the breads were used, the total output in every case, and the faecal output in thirteen out of the fifteen cases, exceeded the intake. The results therefore indicate that riboflavin was synthesized in the intestinal tracts of the children. Since the faecal output exceeded the intake, the values obtained for the apparent percentage absorption of riboflavin from the gastro-intestinal tract were negative.

The levels of faecal excretion of riboflavin at Vohwinkel were distinctly lower than those of children eating the same breads at Duisburg. The presence of more fat in the diets at Vohwinkel than in those at Duisburg may have been responsible, at least in part, for this difference, since there is evidence that in the rat the intestinal synthesis of riboflavin is retarded when the amount of fat in the diet is increased (Czaczkes and Guggenheim, 1946). These authors also found evidence that a high protein diet may reduce intestinal synthesis of riboflavin in the rat, but it seems unlikely that dietary protein was an important factor in the present experiments, since the intakes of protein were somewhat lower at Vohwinkel than at Duisburg (see p. 17). The type of carbohydrate in the diets may possibly have had some effect (see Mannering, Orsini and Elvehjem, 1944). Owing to the lower levels of faecal excretion at Vohwinkel, the balances were positive.

A comparison between the results at the two orphanages shows that the urinary excretion of riboflavin was lower at Vohwinkel than at Duisburg, but that the intakes at the two orphanages were much the same. Since the faecal excretion of riboflavin was also lower at Vohwinkel than at Duisburg, the results suggest that some of the riboflavin synthesized in the intestinal tracts of the children at Duisburg may have been absorbed and utilized. However, it does not seem likely that a very large amount of synthesized riboflavin was absorbed, since the urinary excretion did not represent an unduly high proportion of the intake.

NICOTINIC ACID

Intake

The estimates shown in Tables 8–11, which are based on the total acid-hydrolysable derivatives, were higher for all children than the allowance of 4·7–5·0 mg. per 1,000 total Calories recommended by the U.S. National Research Council (1948) for children aged from 13 to 15 years. In most cases slightly lower intakes would have been recorded if the microbiological values for nicotinic acid in the flours had been used. Owing to the limitations of present knowledge (see p. 95) definite conclusions cannot be reached, but there seems to be no reason to suspect that any of the children's diets were deficient in this vitamin.

TABLE 8

Duisburg: Average daily intake and excretion of nicotinic acid derivatives

Extraction rate of flour (per cent)	Name	Intake (mg. per day) (†)			Intake (mg. per 1,000 total Calories)	Urinary excretion			Faecal (†) excretion (mg. per day) (F)	$\left(\frac{I-F}{I}\right) \times 100$
		Bread and flour	Other foods	Total (I)		Nicotinic acid, acid-hydrolysable derivatives (mg. per day)	N-methylnicotinamide (mg. per day)	Per cent (*) of intake		
100	Hilde	26·4	6·2	32·6	14·2	0·7	10·8	32	20·5	37
	Bernhard	30·4	9·7	40·1	16·3	0·8	3·0	9	17·8	56
	Erika	26·9	8·1	35·0	13·5	1·2	7·1	22	12·6	64
	Average	27·9	8·0	35·9	14·7	0·9	7·0	20	17·0	53
85	Helga	11·3	6·2	17·5	10·8	0·9	5·8	35	4·1	77
	Horst	14·5	9·2	23·7	10·0	1·0	3·0	16	8·3	65
	Agnes	14·0	8·1	22·1	8·7	1·1	5·6	28	5·8	74
	Average	13·3	7·8	21·1	9·8	1·0	4·8	25	6·1	71
70	Marianne	7·1	6·2	13·3	6·9	0·6	5·0	38	2·9	78
	Helmut	14·5	9·7	24·2	9·3	0·6	5·2	22	2·4	90
	Irmtraud	9·1	8·1	17·2	6·7	0·9	5·4	34	2·4	86
	Average, excluding 'Helmut'	8·1	7·2	15·3	6·8	0·8	5·2	36	2·7	82
70 enriched to 100	Hannelore	22·7	7·4	30·1	13·8	1·0	9·4	32	7·7	74
	Hans	30·2	9·7	39·9	14·4	1·2	9·5	25	13·4	66
	Ursula	23·2	8·1	31·3	13·7	1·3	10·4	34	1·3	96
	Average	25·4	8·4	33·8	14·0	1·2	9·8	30	7·5	78
70 enriched to 85	Hanni	11·2	6·2	17·4	11·5	0·8	7·3	43	1·9	89
	Günther	18·0	9·7	27·7	10·9	0·7	4·4	17	2·5	91
	Frieda	17·0	8·1	25·1	9·5	1·1	8·0	33	2·5	90
	Average	15·4	8·0	23·4	10·6	0·9	6·6	29	2·3	90

(*) Sum of acid-hydrolysable derivatives and N-methylnicotinamide, expressed as nicotinic acid.
(†) Expressed as nicotinic acid.

Urinary Excretion

The amounts of acid-hydrolysable derivatives of nicotinic acid excreted were low if they are compared with the values of 1–3 mg. per day reported by Perlzweig *et al.* (1940) for normal adults. The amounts of N-methylnicotinamide excreted were in all cases within the range reported by Huff and Perlzweig (1947) for 67 normal male medical students (3–17, mean 7, mg. per day). Judging by the levels of excretion of N-methylnicotinamide, which up till recently has been regarded as the best available index of nicotinic acid status, it appears probable that in both experiments the children of all groups received adequate supplies of nicotinic acid.

Knox and Grossman (1946, 1947) showed that N-methylnicotinamide is capable of being oxidized *in vivo* to N-methyl-2-pyridone-5-carboxylamide, and they succeeded in isolating the latter compound from human urine after the ingestion of nicotinamide. A method has been worked out (Holman and de Lange, 1949) for the determination of N-methyl-2-pyridone-5-carboxylamide

TABLE 9

Duisburg : Comparison of the intake and output of B-vitamins

Extraction rate of flour (per cent)	Name	Aneurin (mg. per day)			Riboflavin (mg. per day)				Nicotinic acid derivatives (mg. per d…		
		Intake	Output	Balance	Intake	Output		Balance	Intake	Output	Bal…
						Total	Faecal				
100	Hilde	2·17	1·19	+0·98	1·16 (1·7)*	2·54	2·30	−1·38 (−0·8)*	32·6	30·9	+
	Bernhard	2·28	1·15	+1·13	1·44 (1·9)	3·09	2·95	−1·65 (−1·2)	40·1	21·3	+1
	Erika	2·25	1·44	+0·81	1·35 (1·9)	2·80	2·48	−1·45 (−0·9)	35·0	20·2	+1
	Average	2·24	1·26	+0·98	1·32 (1·8)	2·81	2·58	−1·49 (−1·0)	35·9	24·2	+11
85	Helga	1·53	1·04	+0·49	0·61 (1·1)	1·40	1·14	−0·79 (−0·3)	17·5	10·2	+ 7
	Horst	1·81	1·04	+0·77	1·11 (1·6)	1·89	1·63	−0·78 (−0·3)	23·7	12·0	+11
	Agnes	2·22	1·23	+0·99	0·99 (1·6)	1·72	1·48	−0·73 (−0·1)	22·1	11·9	+10
	Average	1·86	1·11	+0·75	0·90 (1·4)	1·67	1·42	−0·77 (−0·3)	21·1	11·4	+ 9
70	Marianne	0·97	0·55	+0·42	0·45 (0·9)	1·35	1·24	−0·90 (−0·5)	13·3	8·0	+ 5
	Helmut	1·36	0·40	+0·96	1·00 (1·3)	1·61	1·41	−0·61 (−0·3)	24·2	7·7	+16
	Irmtraud	1·55	0·70	+0·85	0·75 (1·2)	1·84	1·74	−1·09 (−0·6)	17·2	8·2	+ 9
	Average, excluding 'Helmut'	1·27	0·63	+0·64	0·60 (1·1)	1·60	1·49	−1·00 (−0·4)	15·3	8·2	+ 7
70 enriched to 100	Hannelore	1·96	1·19	+0·77	0·91 (1·3)	1·40	1·12	−0·49 (−0·1)	30·1	17·2	+12
	Hans	2·08	2·01	+0·07	1·13 (1·7)	2·09	1·76	−0·96 (−0·4)	39·9	23·2	+16
	Ursula	2·01	0·87	+1·14	0·83 (1·4)	1·76	1·53	−0·93 (−0·4)	31·3	12·0	+19
	Average	2·02	1·36	+0·66	0·95 (1·5)	1·75	1·47	−0·80 (−0·3)	33·8	17·5	+16
70 enriched to 85	Hanni	1·36	1·05	+0·31	0·61 (0·7)	1·21	1·02	−0·60 (−0·5)	17·4	9·3	+ 8
	Günther	1·99	0·89	+1·10	0·94 (1·1)	1·67	1·42	−0·73 (−0·6)	27·7	7·2	+20
	Frieda	2·13	1·33	+0·80	1·00 (1·1)	1·64	1·24	−0·64 (−0·5)	25·1	10·8	+14
	Average	1·83	1·09	+0·74	0·85 (1·0)	1·51	1·23	−0·66 (−0·5)	23·4	9·1	+14

* Values calculated on the basis of the results obtained for the riboflavin contents of the breads by the rat-growth method shown in brackets.

in human urine. It was not ready in time to be applied to the urines in the Duisburg experiment, but was used in the metabolic studies at Vohwinkel. The results show that considerable amounts of this substance were excreted. Only in the case of one of the children eating unenriched white bread (Hans) was the level of excretion lower than the values observed in healthy adults (cf. Holman and de Lange, 1949; Perlzweig, Rosen, Pearson, Peck and Sparks, 1950). Evidence that this pyridone is the principal metabolite of nicotinic acid in man has been reported by Holman and de Lange (1950b).

Faecal Excretion

In the Duisburg experiment the faecal excretion was highest in the children eating wholemeal bread and the apparent absorption of nicotinic acid from the gastro-intestinal tract tended to be lowest for these children. In none of the children studied did the total output of metabolites of nicotinic acid other than the pyridone, when expressed as nicotinic acid, exceed the intake, even when allowance was made for the possible presence of N-methylnicotinamide in the faeces (Table 1, p. 96).

TABLE 10

Vohwinkel: Average daily intake and excretion of B-vitamins

Extraction rate of flour (per cent)	Name	Intake (mg. per day)			Intake (mg. per 1,000 Cals.)		Urinary excretion					Faecal excretion (mg. per day) (F)	$\left(\frac{I-F}{I}\right) \times 100$
		Bread and flour	Other foods	Total (I)	Total Cals.	Non-fat Cals.	mg. per day			per cent of intake			
Aneurin													
100	Alvin	0·86	0·78	1·64	0·58	0·90	0·46			28		0·38	77
	Willi	0·82	0·71	1·53	0·56	0·87	0·55			36		0·67	56
	Average	0·84	0·75	1·59	0·57	0·89	0·51			32		0·53	67
70	Heinz	0·34	0·74	1·08	0·40	0·61	0·18			17		0·23	79
	Hans	0·29	0·75	1·04	0·41	0·63	0·19			18		0·33	68
	Average	0·32	0·75	1·07	0·41	0·62	0·19			18		0·28	74
70 enriched to 100	Klaus	0·82	0·73	1·55	0·54	0·83	0·45			29		0·16	90
	Karl-Heinz	0·82	0·79	1·61	0·56	0·85	0·35			22		0·24	85
	Average	0·82	0·76	1·58	0·55	0·84	0·40			25		0·20	87
Riboflavin													
100	Alvin	0·42	0·93	1·35	0·48	—	0·06			4		0·45	67
	Willi	0·40	0·93	1·33	0·48	—	0·07			5		0·72	46
	Average	0·41	0·93	1·34	0·48	—	0·07			5		0·59	56
70	Heinz	0·12	0·90	1·02	0·38	—	0·08			8		0·52	49
	Hans	0·11	0·90	1·01	0·40	—	0·05			5		0·65	36
	Average	0·12	0·90	1·02	0·39	—	0·07			7		0·59	42
70 enriched to 100	Klaus	0·24	0·84	1·08	0·38	—	0·21			19		0·62	43
	Karl-Heinz	0·25	0·84	1·09	0·38	—	0·13			12		0·74	32
	Average	0·25	0·84	1·09	0·38	—	0·17			16		0·68	38
Nicotinic acid derivatives							(a)	(b)	(c)	(d)	(e)		
100	Alvin	14·5	12·5	27·0	9·5	—	1·2	5·7	6·0	23	41	5·1	81
	Willi	13·5	12·9	26·4	9·6	—	1·1	6·4	6·6	26	46	7·0	73
	Average	14·0	12·7	26·7	9·6	—	1·2	6·1	6·3	25	44	6·1	77
70	Heinz	6·3	10·1	16·4	6·1	—	1·2	4·5	7·0	32	67	8·5	48
	Hans	5·2	11·5	16·7	6·6	—	1·1	3·7	2·9	26	40	8·5	49
	Average	5·8	10·8	16·6	6·4	—	1·2	4·1	5·0	30	54	8·5	49
70 enriched to 100	Klaus	14·1	12·8	26·9	9·4	—	1·5	6·4	12·9	27	66	8·3	69
	Karl-Heinz	14·0	12·3	26·3	9·1	—	1·0	6·5	11·8	26	63	8·8	67
	Average	14·1	12·6	26·7	9·3	—	1·3	6·5	12·4	27	64	8·6	68

(a) Acid-hydrolysable derivatives of nicotinic acid.
(b) N-methylnicotinamide.
(c) N-methyl-2-pyridone-5-carboxylamide.
(d) Percentage of intake excreted as sum of (a) and (b), expressed as nicotinic acid.
(e) Percentage of intake excreted as sum of (a), (b) and (c), expressed as nicotinic acid.

At Vohwinkel the faecal excretions of nicotinic acid by the children on wholemeal bread were distinctly lower than the levels found in the corresponding group at Duisburg. It is interesting to observe (Table 11) that the amounts of

TABLE 11

Vohwinkel: Comparison of the intake and output of B-vitamins

Extraction rate of flour (per cent)	Name	Intake (mg. per day)	Output (mg. per day)		Balance (mg. per day)	
Aneurin						
100	Alvin	1·64	0·84		+ 0·80	
	Willi	1·53	1·22		+ 0·31	
	Average	1·59	1·04		+ 0·55	
70	Heinz	1·08	0·41		+ 0·67	
	Hans	1·04	0·52		+ 0·52	
	Average	1·07	0·47		+ 0·60	
70 enriched to 100	Klaus	1·55	0·61		+ 0·94	
	Karl-Heinz	1·61	0·59		+ 1·02	
	Average	1·58	0·60		+ 0·98	
Riboflavin						
100	Alvin	1·35	0·51		+ 0·84	
	Willi	1·33	0·79		+ 0·54	
	Average	1·34	0·66		+ 0·68	
70	Heinz	1·02	0·60		+ 0·42	
	Hans	1·01	0·70		+ 0·31	
	Average	1·02	0·66		+ 0·36	
70 enriched to 100	Klaus	1·08	0·83		+ 0·25	
	Karl-Heinz	1·09	0·87		+ 0·22	
	Average	1·09	0·85		+ 0·24	
Nicotinic acid derivatives			(*a*)	(*b*)	(*a*)	(*b*)
100	Alvin	27·0	11·4	16·3	+ 15·6	+ 10·7
	Willi	26·4	13·9	19·2	+ 12·5	+ 7·2
	Average	26·7	12·8	17·9	+ 13·9	+ 8·8
70	Heinz	16·4	13·8	19·5	+ 2·6	— 3·1
	Hans	16·7	12·9	15·2	+ 3·8	+ 1·5
	Average	16·6	13·4	17·5	+ 3·2	— 0·9
70 enriched to 100	Klaus	26·9	15·6	26·0	+ 11·3	+ 0·9
	Karl-Heinz	26·3	15·7	25·3	+ 10·6	+ 1·0
	Average	26·7	15·7	25·8	+ 11·0	+ 0·9

(*a*) Excluding urinary excretion of pyridone.
(*b*) Including urinary excretion of pyridone.

N-methyl-2-pyridone-5-carboxylamide, which were excreted in the urine of the children at Vohwinkel, were sufficiently large to have an important influence on the results of the balances. If the pyridone is excluded from the results, the balances are all positive; when it is included, the balances are in most cases either negative or only slightly positive.

The results provide no evidence as to whether nicotinic acid was synthesized by intestinal micro-organisms or not, since the faecal excretion did not exceed the intake. It is probable that the children were able to derive extra-dietary

supplies of nicotinic acid from ingested tryptophan, but existing experimental evidence suggests that the site of this conversion is in the tissues rather than in the intestinal tract (see Hundley, 1949; Snyderman *et al.*, 1949; Holman and de Lange, 1950c).

Absorption by the Children of the B-vitamins in Wholemeal Bread

At both orphanages the levels of intake and the urinary excretion of aneurin were much the same whether the children were obtaining the vitamin from wholemeal bread or enriched white bread. The results therefore suggest that the aneurin naturally present in the wholemeal bread was utilized as fully as the synthetic aneurin added to the white bread.

In the case of riboflavin and nicotinic acid, however, there were indications that the children eating wholemeal bread tended to excrete less of the vitamin or its metabolites in the urine than the children receiving a similar intake from enriched white bread. The evidence was strongest at Vohwinkel, where the children eating wholemeal bread excreted one third to one half as much riboflavin and N-methyl-2-pyridone-5-carboxylamide in the urine as the children eating enriched white bread. At Duisburg the urinary excretion of the pyridone was unfortunately not determined, but there was a tendency for the amounts of the other metabolites of nicotinic acid excreted to be lower for children eating wholemeal bread than for those having enriched white bread. The urinary excretion of riboflavin by children eating wholemeal bread was also relatively low at Duisburg, but the effect was much less marked than at Vohwinkel, possibly because the Duisburg children were able to utilize some of the riboflavin synthesized in their gastro-intestinal tracts.

These results suggest that the riboflavin and nicotinic acid in the wholemeal bread, or at any rate the substances measured by the chemical and microbiological methods of assay which were employed, were less readily absorbed and utilized by the children than the synthetic riboflavin and nicotinic acid added to the white flour. There is no doubt that other substances, e.g. calcium, may be absorbed very badly from diets consisting largely of wholemeal flour (McCance and Walsham, 1948).

Flour of 85 per cent extraction was not studied in the Vohwinkel experiment. The results at Duisburg showed no clear evidence that the B-vitamins in flour of this degree of extraction were less well utilized than those in the corresponding enriched white flour.

Comparison of the Children's Diets with Average British Food Supplies

Exact figures for the B-vitamin intakes of adults and children in Great Britain, based on direct chemical analysis of the foods eaten, are unfortunately not available. Use has therefore been made of the estimates given by the Special Joint Committee of the Combined Food Board (1944) and the Ministry of Food (1947; 1949) of the amounts of B-vitamins and calories furnished daily per head of population by the food supplies moving into civilian consumption in the United Kingdom. These estimates represent the average vitamin supplies of all age-groups, and make no allowance for food wastage. They cannot therefore be compared directly with the German children's daily vitamin intakes. Moreover, it should be noted that the estimates are based on selected values for the concentrations of B-vitamins in the various foods, and that they do not take into account the losses of the vitamins in the cooking of foods.

In the case of the orphanage diets, detailed information on B-vitamin intakes is available only for the children who took part in the metabolic studies, during the particular week when these studies were made. It seems probable, however, that the results are truly representative, both because comparable levels of urinary excretion were found for other children in the same bread groups at other times, and also because calculations of the amounts of the vitamins in the average diet of all the children over the whole year (see p. 16) gave similar results to those found by analysis.

In Table 12 the amounts of aneurin and riboflavin in relation to calories furnished by the children's whole diets and by the chief classes of foods comprising these diets are compared with the corresponding British supplies of the vitamins. The comparison has not been extended to include nicotinic acid, in

TABLE 12

B-vitamin composition of the main classes of foods in the German children's diets and in the British food supplies, expressed in relation to their energy value

Diet	Aneurin (mg. per 1,000 non-fat Calories) contained in:				Riboflavin (mg. per 1,000 total Calories) contained in:			
	Grain products (1)	Sugar, syrups, fats and oils (2)	Foods other than 1 and 2	All foods	Grain products (1)	Sugar, syrups, fats and oils (2)	Foods other than 1 and 2	All foods
*Duisburg diets**								
Containing unenriched flour of 70 per cent extraction	0·33–0·46	nil	1·48–2·46	0·55–0·65	0·11–0·18	nil	0·68–1·15	0·23–0·29
Containing flour of 85 per cent extraction ..	0·77–0·95	nil	1·52–2·50	0·83–1·06	0·28–0·39	nil	0·71–1·12	0·38–0·47
Containing flour of 100 per cent extraction ..	0·87–1·00	nil	1·52–2·39	0·95–1·04	0·48–0·54	nil	0·71–1·05	0·51–0·58
*Vohwinkel diets**								
Containing unenriched flour of 70 per cent extraction	0·38–0·40	nil	1·13–1·17	0·61–0·63	0·14	nil	1·21–1·23	0·38–0·40
Containing flour of 100 per cent extraction ..	1·03–1·06	nil	1·03–1·14	0·87–0·90	0·47–0·49	nil	1·17–1·18	0·48
Foods available for consumption in U.K.†								
Pre-war	0·25	nil	1·98	0·66	0·12	nil	1·31	0·53
1941	0·46	nil	1·97	0·75	0·21	nil	1·41	0·57
1943	0·92	nil	1·95	1·06	0·45	nil	1·53	0·73
1945	—	—	—	0·95	—	—	—	0·61
1947	—	—	—	0·94	—	—	—	0·69

* Values observed during the metabolic studies.
† Estimates made by the Special Joint Committee of the Combined Food Board (1944) and by the Ministry of Food (1947; 1949).

view of the limited state of present knowledge of its metabolism. From an examination of the values for 'all foods' it appears probable that the children's diets at the orphanages, as well as the British food supplies in each of the years studied from before the war up to 1947, provided adequate amounts of aneurin (see pp. 104, 105). The amounts of riboflavin in the food supplies of the United Kingdom were adequate judging by existing evidence (see p. 107), but the amounts available to the children receiving unenriched white bread were

relatively low. Further examination of the table shows that the figures for aneurin in the unenriched white bread used in the feeding experiments are somewhat higher than the corresponding value given for the grain products available in the United Kingdom before the war, but the latter, which equals the lowest reported value for aneurin in flour of 70 per cent extraction, may not be a true average for all the grain products in use at that time. In the case of foods other than grain products, sugar and fats, the figures for the children's diets are in most cases a little lower than the corresponding values for the British supplies, but too close a comparison is probably not warranted, in view of the considerations mentioned above.

Apart from variations in flour extraction, the chief differences between the experimental diets and the British food supplies lay, not in the composition of the foods, but in the proportions of the total diet which they represented. This is borne out by the figures given in Table 13, which show that grain products

TABLE 13

Proportions of the total B-vitamins and calories in the German children's diets and in the British food supplies which were contributed by various classes of foods

Diet	Percentage of total aneurin contributed by:			Percentage of total riboflavin contributed by:			Percentage of total calories contributed by:		
	Grain products (1)	Sugar, syrups, fats and oils (2)	Foods other than 1 and 2	Grain products (1)	Sugar, syrups, fats and oils (2)	Foods other than 1 and 2	Grain products (1)	Sugar, syrups, fats and oils (2)	Foods other than 1 and 2
*Duisburg diets**									
Containing unenriched flour of 70 per cent extraction	46–50	nil	50–54	33–41	nil	59–67	68–79	7–10	14–25
Containing flour of 85 per cent extraction ..	65–74	nil	26–35	51–64	nil	36–49	69–77	7–11	15–25
Containing flour of 100 per cent extraction ..	66–78	nil	22–34	63–74	nil	26–37	69–79	7– 8	13–24
*Vohwinkel diets**									
Containing unenriched flour of 70 per cent extraction	28–31	nil	69–72	11–12	nil	88–89	31–32	40	28–29
Containing flour of 100 per cent extraction ..	52–54	nil	46–48	30–31	nil	69–70	30–31	41	28–29
Foods available for consumption in U.K.†									
Pre-war	18	nil	82	7	nil	93	30	32	38
1941	36	nil	64	15	nil	85	39	27	34
1943	48	nil	52	23	nil	77	37	26	37
1945	—	—	—	—	—	—	38	25	37
1947	—	—	—	—	—	—	36	26	38

* Values observed during the metabolic studies.
† Estimates made by the Special Joint Committee of the Combined Food Board (1944) and by the Ministry of Food (1947; 1949).

furnished twice as high a proportion of the total calories in the Duisburg diet as in the British food supplies, while sugar and fats, the only classes of foods which are devoid of B-vitamins, accounted for considerably more calories in the Vohwinkel diet than in the British food supplies. The British supplies contained more foods other than grain products, sugar and fats than did any of the children's diets; since these foods are the richest in B-vitamins, they contributed a large proportion of the B-vitamins in the British supplies.

Summary

1. Preliminary investigations were made into the B-vitamin composition of the experimental flours and breads using various methods of assay. The results indicated that the concentrations of aneurin in the natural flours were in every case within the normal ranges, but that the concentration in the flour of 70 per cent extraction was at the upper limit of the normal range.

2. The nutritional state of the children with respect to aneurin, riboflavin and nicotinic acid was investigated by determining the levels of intake and of urinary and faecal excretion of the vitamins and their metabolites at various stages of the experiments.

3. Owing to limitations of present knowledge about the exact amounts of physiologically available B-vitamins in cereals, particularly of riboflavin and nicotinic acid, the levels of urinary excretion probably afforded a more accurate index of B-vitamin status than did estimates of the dietary intake. There was, however, good agreement between the main conclusions reached by these two independent methods of investigation.

4. The results showed no evidence that any of the children's diets were deficient in the vitamins, but the margin of reserve appeared to be small in the case of the children eating unenriched white bread. The supplies of riboflavin were probably nearest to the margin of safety.

5. The results of the Duisburg experiment indicated that riboflavin was synthesized in the gastro-intestinal tracts of children receiving all the five sorts of bread. It is not possible to state with certainty whether or not synthesized vitamins were absorbed and utilized by the children, but there were some indications that the Duisburg children may have been able to utilize synthesized riboflavin. If the children eating unenriched white bread were obtaining extra amounts of B-vitamins in this way, they might well have shown no signs of deficiency on an even lower B-vitamin intake.

6. The results provided some indications that children eating wholemeal bread excreted less riboflavin and metabolites of nicotinic acid in their urine than did children on enriched white bread. This observation suggests that the riboflavin and nicotinic acid in whole-wheat flour may not be freely absorbed by children.

References

Alexander, B. and Landwehr, G. (1946). Studies of thiamine metabolism in man. 1. Thiamine balance. The normal requirement of vitamin B_1 and the role of fecal thiamine in human nutrition. *J. clin. Invest.*, **25**, 287.

Andrews, J. S. (1943). Report of 1942–43 methods of analysis sub-committee on riboflavin assay. *Cereal Chem.*, **20**, 613.

Benson, R. A., Witzberger, C. M. and Slobody, L. B. (1943). An evaluation of the blood and urinary thiamine determinations in vitamin B_1 sub-nutrition. *J. Pediat.*, **23**, 437.

Caro, de L. and Rindi, G. (1950). Sparing action of fats and pyruvate in the blood of rats in avitaminosis B_1. *Nature*, **167**, 114.

Chaudhuri, D. K. and Kodicek, E. (1950a). Purification of a precursor of nicotinic acid from wheat bran. *Nature*, **165**, 1022.

Chaudhuri, D. K. and Kodicek, E. (1950b). The biological activity for the rat of a bound form of nicotinic acid present in bran. *Biochem. J.*, **47**, xxxiv.

Copping, A. M. (1946). Factors affecting the nutritive value of bread as human food. Vitamin values of different types of flour. *Proc. Nutr. Soc.*, **4**, 9.

Czaczkes, J. W. and Guggenheim, K. (1946). The influence of diet on the riboflavin metabolism of the rat. *J. biol. Chem.*, **162**, 267.

Dann, W. J. and Handler, P. (1941). The quantitative estimation of nicotinic acid in animal tissues. *J. biol. Chem.*, **140**, 201.

Davis, M. V., Oldham, H. G. and Roberts, L. J. (1946). Riboflavin excretions of young women on diets containing varying levels of the B vitamins. *J. Nutrit.*, **32**, 143.

DENKO, C. W., GRUNDY, W. E., WHEELER, N. C., HENDERSON, C. R. and BERRYMAN, G. H. (1946). The excretion of B-complex vitamins by normal adults on a restricted intake. *Arch. Biochem.*, **11**, 109.

ELLINGER, P. (1947). The fate of nicotinamide methochloride and the effect of liver poisons on its elimination rate in the rat. *Biochem. J.*, **41**, 308.

GREENBERG, L. D. and RINEHART, J. F. (1945). Methods for determination of thiamine in blood and tissues with observations on relative contents. *Proc. Soc. exp. Biol. Med.*, **59**, 9.

HATHAWAY, M. L. and LOBB, D. E. (1946). A comparison of riboflavin synthesis and excretion in human subjects on synthetic and natural diets. *J. Nutrit.*, **32**, 9.

HOLMAN, W. I. M. (1946). The amounts of vitamin B_1 in cereals and the extent to which they supply human requirements in various dietaries. *Nutr. Abstr. Rev.*, **15**, 387.

HOLMAN, W. I. M. and DE LANGE, D. J. (1949). Methods for the determination of N-methyl-2-pyridone-5-carboxylamide and of N-methyl-2-pyridone-3-carboxylamide in human urine. *Biochem. J.*, **45**, 559.

HOLMAN, W. I. M. and DE LANGE, D. J. (1950a). Significance of tryptophane in human nicotinic acid metabolism. *Nature*, **165**, 112.

HOLMAN, W. I. M. and DE LANGE, D. J. (1950b). Metabolism of nicotinic acid and related compounds by humans. *Nature*, **165**, 604.

HOLMAN, W. I. M. and DE LANGE, D. J. (1950c). Role of tryptophane and other amino acids in the metabolism of nicotinic acid by humans. *Nature*, **166**, 468.

HUFF, J. W. and PERLZWEIG, W. A. (1947). The fluorescent condensation product of N^1-methylnicotinamide and acetone. II. A sensitive method for the determination of N^1-methylnicotinamide in urine. *J. biol. Chem.*, **167**, 157.

HUNDLEY, J. M. (1949). Influence of intestinal bacteria on synthesis of nicotinic acid from tryptophane. *Proc. Soc. exp. Biol. Med.*, **70**, 592.

KEYS, A., HENSCHEL, A. F., MICKELSEN, O. and BROZEK, J. M. (1943). The performance of normal young men on controlled thiamine intakes. *J. Nutrit.*, **26**, 399.

KEYS, A., HENSCHEL, A., TAYLOR, H. L., MICKELSEN, O. and BROZEK, J. (1945). Experimental studies on man with a restricted intake of the B vitamins. *Amer. J. Physiol.*, **144**, 5.

KLEIN, J. R., PERLZWEIG, W. A. and HANDLER, P. (1942). Determination of nicotinic acid in blood cells and plasma. *J. biol. Chem.*, **145**, 27.

KNOX, W. E. and GROSSMAN, W. I. (1946). A new metabolite of nicotinamide. *J. biol. Chem.*, **166**, 391.

KNOX, W. E. and GROSSMAN, W. I. (1947). The isolation of the 6-pyridone of N^1-methylnicotinamide from urine. *J. biol. Chem.*, **168**, 363.

KODICEK, E. (1946). Factors affecting the nutritive value of bread as human food. *Proc. Nutr. Soc.*, **4**, 22.

MCCANCE, R. A. and WALSHAM, C. M. (1948). The digestibility and absorption of the calories, proteins, purines, fat and calcium in wholemeal wheaten bread. *Brit. J. Nutr.*, **2**, 26.

MANNERING, G. J., ORSINI, D. and ELVEHJEM, C. A. (1944). Effect of the composition of the diet on the riboflavin requirement of the rat. *J. Nutrit.*, **28**, 141.

MASON, H. L. and WILLIAMS, R. D. (1942). The urinary excretion of thiamine as an index of the nutritional level: assessment of the value of a test dose. *J. clin. Invest.*, **21**, 247.

MINISTRY OF FOOD (1947). Food consumption levels in United Kingdom. H.M. Stationery Office, London.

MINISTRY OF FOOD (1949). Food consumption levels in United Kingdom. H.M. Stationery Office, London.

MORELL, D. B. and SLATER, E. C. (1946). The fluorimetric determination of riboflavin in urine. *Biochem. J.*, **40**, 652.

NAJJAR, V. A. and KETRON, K. C. (1944). An improved thiochrome method for the determination of thiamine in urine. *J. biol. Chem.*, **152**, 579.

NUTRITION REVIEWS (1947a). Riboflavin and human nutrition. *Nutr. Rev.*, **5**, 60.

NUTRITION REVIEWS (1947b). A new metabolite of niacin. *Nutr. Rev.*, **5**, 223.

NUTRITION REVIEWS (1950). Influence of thiamine intake on physiologic responses. *Nutr. Rev.*, **8**, 45.

PERLZWEIG, W. A., LEVY, E. D. and SARETT, H. P. (1940). Nicotinic acid derivatives in human urine and their determination. *J. biol. Chem.*, **136**, 729.

PERLZWEIG, W. A., ROSEN, F., PEARSON, P. B., PECK, B. J. and SPARKS, P. (1950). Comparative studies in niacin metabolism. The fate of niacin in man, rat, dog, pig, rabbit, guinea-pig, goat, sheep and calf. *J. Nutrit.*, **40**, 453.

RITTER, E. DE, MOORE, M. E., HIRSCHBERG, E. and RUBIN, S. H. (1949). Critique of methods for the determination of riboflavin in urine. *J. biol. Chem.*, **175**, 883.

SARETT, H. P. (1943). A direct method for the determination of N-methyl derivatives of nicotinic acid in urine. *J. biol. Chem.*, **150**, 159.

SLATER, E. C. and MORELL, D. B. (1946a). A modification of the fluorimetric method of determining riboflavin in biological materials. *Biochem. J.*, **40**, 644.

SLATER, E. C. and MORELL, D. B. (1946b). The effect of storage on the riboflavin content of urine. *Austral. exp. Biol. med. Sci.*, **24**, 121.

SNYDERMAN, S. E., KETRON, K. C., CARRETERO, R. and HOLT, L. E. (Jr.) (1949). Site of conversion of tryptophane into nicotinic acid in man. *Proc. Soc. exp. Biol. Med.*, **70**, 569.

SPECIAL JOINT COMMITTEE OF THE COMBINED FOOD BOARD (1944). Food consumption levels in the United States, Canada and the United Kingdom. H.M. Stationery Office, London.

U.S. NATIONAL RESEARCH COUNCIL (1948). Recommended dietary allowances. *Nat. Res. Coun. Reprint and Circular Series No.* 129.

VITAMIN B_1 SUB-COMMITTEE OF THE ACCESSORY FOOD FACTORS COMMITTEE OF THE MEDICAL RESEARCH COUNCIL AND THE LISTER INSTITUTE (1943). The vitamin B_1 content of national flour and bread—the results of comparative tests by various methods. *Biochem. J.*, **37**, 433.

WILLIAMS, R. D., MASON, H. L., CUSICK, P. L. and WILDER, R. M. (1943). Observations on induced riboflavin deficiency and the riboflavin requirement of man. *J. Nutrit.*, **25**, 361.

WILLIAMS, R. D., MASON, H. L., SMITH, B. F. and WILDER, R. M. (1942). Induced thiamine (vitamin B_1) deficiency and the thiamine requirement of man. *Arch. intern. Med.*, **69**, 721.

WILLIAMS, R. D., MASON, H. L. and WILDER, R. M. (1943). The minimum daily requirement of thiamine of man. *J. Nutrit.*, **25**, 71.

WILLIAMS, R. J., EAKIN, R. E., BEERSTECHER, E. (Jr.) and SHIVE, W. (1950). *The biochemistry of B vitamins.* Reinhold Publishing Corp., New York.

APPENDIX D: THE SKELETAL DEVELOPMENT OF THE CHILDREN AT THE BEGINNING AND END OF THE PERIOD OF EXPERIMENTAL FEEDING

by F. R. Berridge and Kathleen M. Prior

It is now generally recognized that children do not grow normally unless an adequate diet is provided, but the effect of a diet deficient in calories on the ossification of children's bones has never been made clear. It seemed to be of interest, therefore, to find out whether the skeletal development of the children taking part in this investigation was retarded and to assess the progress of ossification of their bones over the year during which they were being given unlimited amounts of the diets already described.

Various radiological methods have been used for the assessment of skeletal development and it is important to consider these briefly in order that the significance of the results obtained in this study may be understood. The radiographic demonstration of the presence of the various centres of ossification or of their union has for long provided criteria upon which skeletal development may be assessed (von Ranke, 1896, 1898; Behrendsen, 1897; Poland, 1898; Pryor, 1906, 1925; Heimann and Potpeschnigg, 1907; Rotch, 1909; Stettner, 1920–21, 1935; Engelbach and McMahon, 1924; Davies and Parsons, 1927; Shelton, 1931; Francis and Werle, 1939; Francis, 1940; Flecker, 1942; Lurie, Levy and Lurie, 1943), but certain disadvantages detract from their use for this purpose. Firstly, there are long periods in the life of a child in which no new centres normally appear or unite; secondly, there are wide variations in the ages at which the centres appear in normal children, and thirdly, the first appearance of ossification in the carpal centres is influenced by the social status and the environment of the child (Stettner, 1920–21). Nevertheless, the radiological demonstration of the appearance and union of the epiphyses is the only purely objective method of assessing ossification of the skeleton. Todd (1932) devised a method of assessing the progress of ossification which was based upon the changes in the architecture of the bones as depicted on radiographs, together with the appearance and union of some of the centres of ossification. His method is not subject to the disadvantages mentioned above, and provides a continuous scale of changes in the radiographic appearance of the bones from birth to maturity. Todd published data for estimating the degree of ossification of the upper and lower extremities (1932), but described and illustrated the changes in the bones of the hand in much greater detail (1937) than those of other regions. Similar methods have been used by other workers (Siegert, 1935; Flory, 1936; Greulich and Pyle, 1950).

An entirely different approach to the problem of assessing skeletal development was employed by Gates (1924) and Freeman and Carter (1924), who measured the areas of all the carpal centres of ossification shown on radiographs of the hands of children of different age groups, and calculated the ratio between the sum of these areas and the 'total carpal area', an area on the radiographs enclosed by lines connecting certain bony points around the carpus. The technical difficulties of this procedure were very great, and Sawtell (1929) devised a much simpler method, based upon the ratio between the maximum width of the lower radial epiphysis and the maximum width of the lower end of the radial diaphysis as shown radiographically. Neither of these ratios, however,

indicates the degree of ossification, but both show the relationship between growth, as indicated by the total carpal area or the width of the lower end of the radius, and ossification, as shown by the areas of the carpal centres or the width of the lower radial epiphysial centre. There are, therefore, several methods of assessing the 'bone age' of a child, and when this term is used it is essential to state the criteria from which it has been estimated.

Material and Radiographic Technique

The bones of three series of children were examined radiologically. Two of the series consisted of the children in orphanages in Duisburg and Vohwinkel taking part in the experiments described in this report, and the third was composed of healthy British schoolchildren of comparable ages who served as controls. The X-ray examinations of the German children were carried out at the Kinderklinik, Duisburg and the X-ray Department of the I.G.-Farbenindustrie, Wuppertal, by kind permission of Professor Thomas and Professor Domagk. The children in the first series comprised 50 girls aged 5–14 years and 56 boys aged 5–13 years in the orphanage at Duisburg. The children in the Vohwinkel orphanage formed the second series, which consisted of 27 girls and 30 boys aged 5–14 years. The bones of both these series of children were X-rayed according to the technique described below, immediately before the experimental diets were started and again after the children had been living on the diets for a period of a year. It will be noted that a few more children were included in this radiological study than are recorded in the Tables on pp. 10, 11 as completing the 12 months of the investigation. It took some weeks to make all the X-ray examinations of the children; those at the beginning were made during the preliminary period, before the experiment started, and the final examinations were made 12 months later, during the last few weeks of the experimental year. Several children, who left before the experiment terminated, were still there when the X-ray examinations were made. One girl at Duisburg who completed the year of investigation missed the final X-ray examination.

The control series of British children was composed of 48 girls and 54 boys aged 5–15 years. They were attending a Junior or a Central Secondary Modern School in Cambridge and may probably be considered representative of healthy girls and boys in this country. Their mean height and weight were up to the American standards of O'Brien, Girshick and Hunt (1941). Their bones were examined only once.

At the beginning of the investigation at Duisburg, radiographs were taken of the left elbow (antero-posterior view) and of the left wrist including the proximal part of the hand (antero-posterior and lateral views) of each child. Three months after the commencement of the experimental diet, films were taken of the wrists of 20 girls aged 5–9 years and 26 boys aged 5–11 years in order to determine whether the new bone laid down round the carpal centres during this period differed in texture from that present before, and to see if any lines of arrested growth attributable to the period of undernutrition (Stettner, 1921; Harris, 1933) were present. At the end of the year, films were taken of the left arm and leg of each child according to the technique of Todd (1932). Thus films of more regions of the body were taken at the end of the investigation than at the beginning of it. This was done because a preliminary survey of the first radiographs revealed some delay in the appearance of the epiphysial centres;

it was thought that the additional films might be of value in comparing the ossification of these children's bones with those of the control children in England. Films were taken of the left arms and legs of the children at the Vohwinkel orphanage, both at the beginning and at the end of the experimental year according to the above technique. The same technique was used in examining the children in England.

The films of the hands and wrists and also of the knees were used to determine the degree of ossification or 'bone age' of each child according to the standards of Todd (1932, 1937), which will be referred to as the American standard. Separate standards for girls and boys are given by Todd for hands (Todd, 1937), but in the case of knees standards for boys only are available (Todd, 1932). The interval of time between each successive standard is 6 months. The bone age of the children was therefore expressed in increments of 6 months, that is, n years 3 months and n years 9 months. The chronological ages were also expressed in 6-monthly increments to correspond with the bone ages; children examined on their birthday or up to 6 months afterwards were placed in the age group n years 3 months, and those examined 6 months or more after their birthday were placed in the age group n years 9 months.

The assessment of the bone age of a child from a film is a subjective procedure; in order to avoid any unconscious bias in estimating bone age from the radiographs the following method was adopted. The films of the hands of all the girls at Duisburg and Vohwinkel, both at the beginning and at the end of the investigations, and also of the British girls, were mixed together, so that when scrutinizing the films the two authors did not know to which set any particular radiograph belonged. The same procedure was used for the films of the boys' hands. Each film was assigned a bone age according to Todd's (1937) standards for girls or boys. All the films of the knees of the Duisburg, Vohwinkel and British girls and boys were similarly mixed, but here Todd's (1932) standards for boys were employed for all the children.

Scatter diagrams for each series of children were drawn to show the relationship between the chronological age and the bone age (a) of the hand, and (b) of the knee. From these, regression lines were calculated and it was found that a straight line could be fixed correlating the bone age with the chronological age in each series. Statistical methods of analysis were applied to these results. The estimation of the precision of the average increase in the bone age over the year was based upon the distribution of the increase for each individual, which gave a much greater accuracy than was possible merely by testing the difference between the average chronological age and the average bone age for the whole series at the beginning and at the end of the year.

To compare the epiphysial development of the German children with that of the British children, the girls and boys in each series were separated into yearly age groups; those examined on their birthday or during the following year were assigned an age group n years. A table was made to show the number of children in each yearly age group for all the series of girls and boys. The radiographs of the extremities of the children were examined for the presence of the centres of ossification. The centres were considered separately, and a table, similar to that constructed for the chronological ages, was made for each one, showing the number of girls and boys in each yearly age group in the three series in whom the centre was ossified. Thus the ages at which the centres of ossification appeared in the different series of children could be compared.

Unfortunately, the number of girls and boys at each age was insufficient for statistical methods to be applied to these results.

Results

THE INITIAL OSSIFICATION OF THE BONES OF THE HANDS AND ITS PROGRESS OVER THE YEAR OF EXPERIMENTAL FEEDING

A summary of the findings for the different bread groups has already been given (p. 41). No differences could be detected between the bone development of the children eating the different kinds of bread, and it has therefore been decided in this more detailed discussion of the children's progress to consider the children in each orphanage as a whole, irrespective of the kind of bread they ate. This has enabled a satisfactory analysis of the results to be made.

In Table 1 the means of the differences between the bone ages of the hands, which may be termed the 'hand ages', and the chronological ages of the British children are set out, together with those of the Duisburg and Vohwinkel

TABLE 1

Means of the differences between the bone ages of the hands and the chronological ages of the British children and of the Duisburg and Vohwinkel children at the beginning and end of the year of experimental diets

(*All bread groups together*)

Children	Girls				Boys			
	No. in sample	Difference between hand age and chronological age Mean (with standard error) (months)		Increase in hand age in excess of that expected during 12 months Mean (with standard error) (months)	No. in sample	Difference between hand age and chronological age Mean (with standard error) (months)		Increase in hand age in excess of that expected during 12 months Mean (with standard error) (months)
		Beginning	End			Beginning	End	
British	48	− 0·4 ±1·5			54	− 3·3 ±1·6		
Duisburg	50	−15·7 ±1·8	−11·0 ±1·7	+4·7 ±0·7	56	−19·6 ±1·8	−12·2 ±1·8	+7·4 ±0·7
Vohwinkel	27	−20·9 ±2·2	−14·7 ±1·8	+6·2 ±0·8	30	−20·6 ±2·4	−14·0 ±2·4	+6·6 ±0·9

children before and after the year of experimental diets. At the beginning the degree of development of the bones of the hands of the girls and boys at the two German orphanages was between 15·3 and 20·5 months behind that of the British children. The bones of the hands were less well developed at Vohwinkel than at Duisburg, which was rather unexpected because, judged by other standards, the nutritional state of the Vohwinkel children was better than that of the Duisburg children (see p. 5). During the 12 months of experiment the bones of both Duisburg and Vohwinkel children matured faster than normal, so that at the end the degree of development was nearer to that of British and American children of similar ages, but it was still about 12 months behind.

Regression lines correlating hand age with chronological age over the whole age range of 5 to 15 years have been constructed (Figs. 1 and 2) to show whether the older or younger children were most retarded as regards their bone development. Before the experiment began the hand ages of the Duisburg girls were significantly below their chronological ages at all ages between 5 and 14 years. The hand ages of the Duisburg boys at this time were also behind their

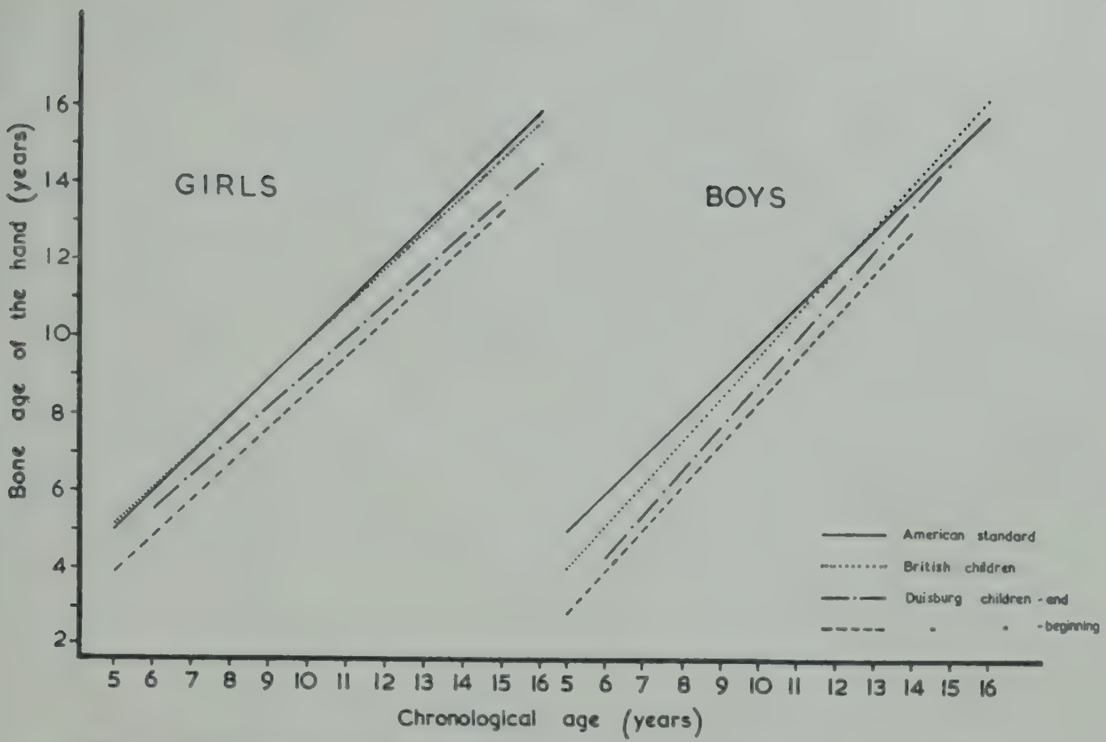

FIG. 1. The relationship between the chronological age and the bone age of the hand for the British children, and for the Duisburg children at the beginning and end of the year of experiment.

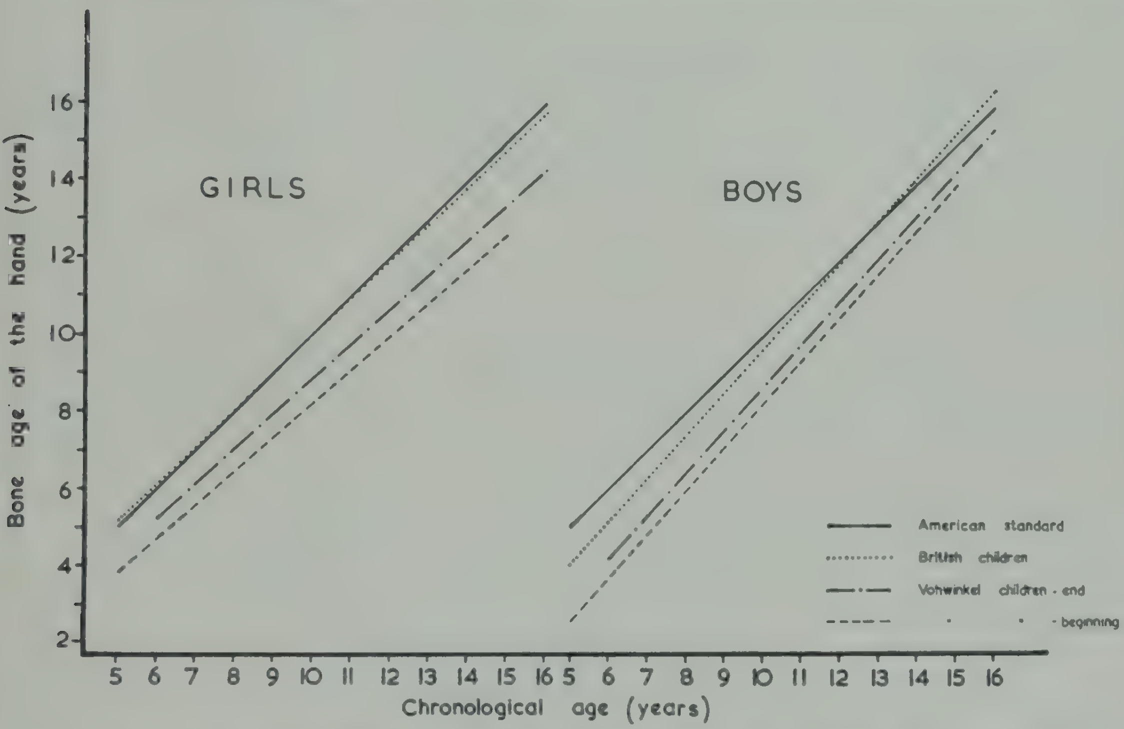

FIG. 2. The relationship between the chronological age and the bone age of the hand for the British children, and for the Vohwinkel children at the beginning and end of the year of experiment.

chronological ages, and the hand ages of the younger boys tended to be relatively lower than those of the older ones. It should be noted that, whereas the mean hand age and the mean chronological age of the British girls were the same at all ages, the mean hand age of the youngest British boys was less than their chronological age, but this difference decreased progressively as they grew older,

and by $12\frac{1}{2}$ years it had disappeared. Why the bones of the hands of the younger American boys should be more mature it is impossible to say, but it is of interest that the regression lines for the British and Duisburg boys are almost parallel; the hand ages of both the Duisburg girls and the Duisburg boys were, on the average, behind those of the British children by the same amount, 16 months. At the end of the year of the experimental diets the bones of the hands of the Duisburg children of all ages had matured by significantly more than 12 months, the girls by 4·7 ± 0·7 months and the boys by 7·4 ± 0·7 months over and above 12 months, but their ossification was still significantly behind that of the British children.

At the beginning of the investigation at the Vohwinkel orphanage the mean hand age of the girls was significantly less than their mean chronological age and the difference was approximately the same at all ages (Fig. 2). The bone development of the Vohwinkel boys was also retarded, but like the British and Duisburg boys, the difference tended to be relatively greater in the younger than in the older ones. After the year of experimental feeding the hand ages of the Vohwinkel girls and boys of all ages increased significantly more than the expected amount, the girls by 6·2 ± 0·8 months and the boys by 6·6 ± 0·9 months over and above 12 months.

A comparison between the radiographs of the wrists of the Duisburg children taken at the beginning of the investigation and those taken 3 months later showed that the trabecular pattern and radiographic opacity of the new bone which had been laid down around the carpal centres during the 3 months was the same as that present before. It would appear, therefore, that the bone originally present had been normal in structure and composition. No transverse lines of arrested growth were observed in the lower ends of the radii of any of these children. Presumably the check in their growth had not been abrupt enough to cause such lines of sclerosis.

COMPARISON OF THE DEGREE OF DEVELOPMENT OF THE BONES OF THE KNEES

Table 2, constructed in the same way as Table 1, shows the means of the differences between the chronological ages and the bone ages of the knees for

TABLE 2

Means of the differences between the bone ages of the knees and the chronological ages of the British children, and of the Duisburg children at the end, and of the Vohwinkel children at the beginning and end of the year of experimental diets (All bread groups together)

Children	Girls: No. in sample	Girls: Difference between bone age of the knee, assessed according to the standards for boys, and chronological age Mean (with standard error) (months) — Beginning	Girls: Difference … — End	Girls: Increase in bone age of the knee in excess of that expected during 12 months Mean (with standard error) (months)	Boys: No. in sample	Boys: Difference between bone age of the knee and chronological age Mean (with standard error) (months) — Beginning	Boys: Difference … — End	Boys: Increase in bo[…] age of the kn[…] in excess of th[…] expected duri[…] 12 months Mean (with standard erro[…]) (months)
British	48	+19·9 ±1·5			54	+ 0·4 ±1·1		
Duisburg	50		+6·3 ±1·4		56		−11·7 ±1·2	
Vohwinkel	27	+ 2·7 ±2·2	+4·0 ±1·9	+1·3 ±0·9	30	−12·4 ±2·1	−11·6 ±2·2	+0·8 ±1·0

the British girls and boys, together with those for the Duisburg children at the end of the period of experimental feeding and those for the Vohwinkel children at the beginning and the end of their investigation. Since girls' bones mature more rapidly than those of boys, and since boys' standards have had to be used for both sexes, the British girls' knees were considerably in advance of the standard, whereas the bone age of the boys' knees agreed with the standard. The ossification of the Duisburg girls' knees at the end of the investigation was in advance of the boys' standard by about 6 months, but it was over a year behind that of the British girls' knees. The Duisburg boys' knees at this time were retarded by just under a year as compared with the American standard and the British mean.

Before the experiment started the knees of the Vohwinkel girls appeared slightly more mature than the American standard for boys' knees, but they were nearly a year and a half behind those of the British girls. The ossification of the Vohwinkel boys' knees was a year behind that of the British boys and the American standard at this time. After the year of experimental feeding the bone age of the knees of both girls and boys increased by just over a year. From Figs. 3 and 4 it will be seen that there was a tendency for the younger girls to have a relatively higher knee age than the older ones, but this was not significant. In the case of the boys at Duisburg, however, the opposite was observed; the younger boys had a relatively lower knee age than the older ones, and this was statistically significant.

At the end of the period of experimental feeding, therefore, the mean bone ages of the hands and knees of the Vohwinkel children were still behind the corresponding mean bone ages of the British children by about a year. The ossification of the knees resembled that of the hands in that it was backward at

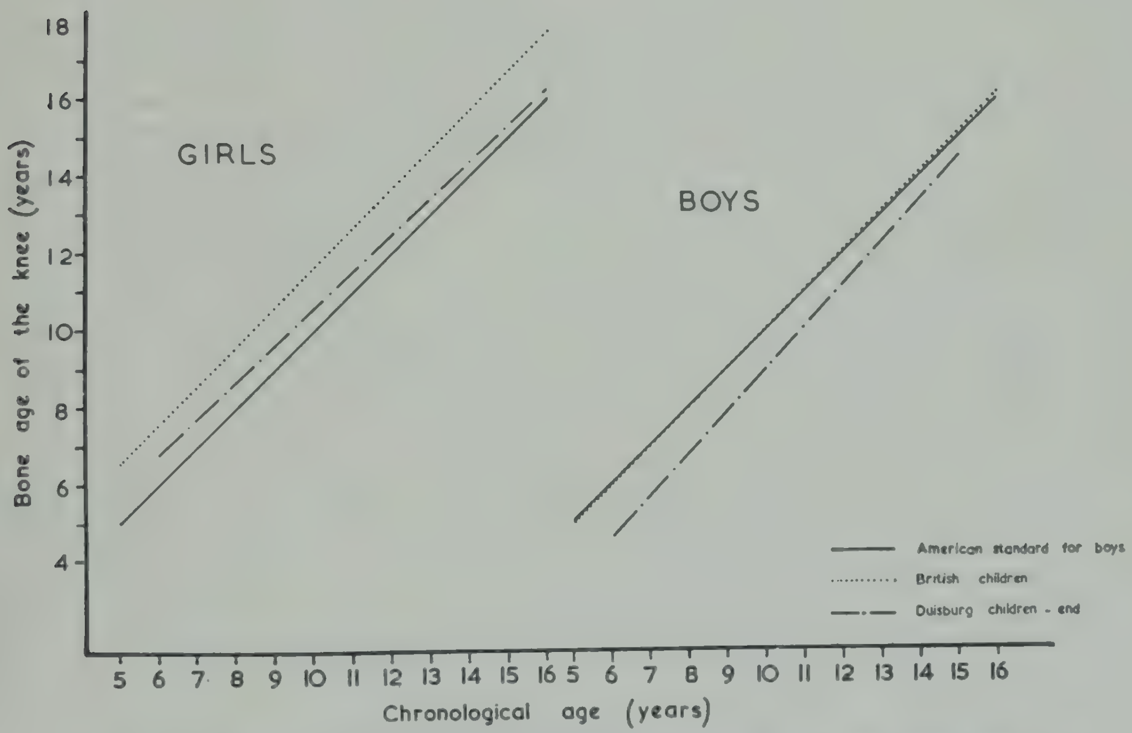

Fig. 3. The relationship between the chronological age and the bone age of the knee for the British children, and for the Duisburg children at the end of the year of experiment.

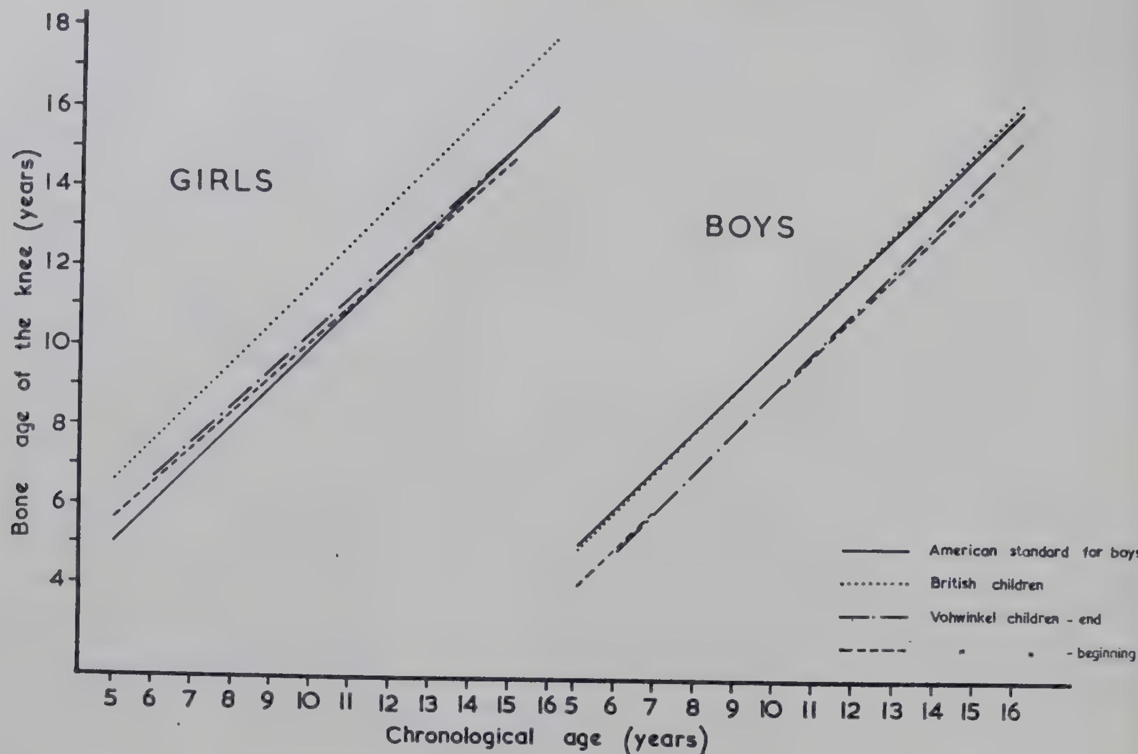

FIG. 4. The relationship between the chronological age and the bone age of the knee for the British children, and for the Vohwinkel children at the beginning and end of the year of experiment.

the start of the investigation but, unlike that of the hands, it did not increase appreciably more than was to be expected during the year of experimental diet. The failure of the bones of the knees of the German children to mature over the year by as large an increment as the bones of the hands is probably more apparent than real, the smaller number of bones in the knees than in the hands and their simpler architecture providing fewer indices for assessing bone age.

COMPARISON OF THE EPIPHYSIAL DEVELOPMENT OF THE BONES

When the epiphysial development of the bones of the German girls and boys was compared with that of the British girls and boys, it was seen that the ossification of the German children was initially 1–2 years behind that of the British children. By the end of the investigation the epiphysial development of the German children had improved a little, the girls slightly more than the boys. The German children, however, still had not attained the degree of epiphysial development observed in the British children.

CORRELATION BETWEEN THE HEIGHTS OF THE GERMAN CHILDREN AND THE OSSIFICATION OF THEIR BONES

So far skeletal development has been considered in relation to chronological age, and it has been shown that ossification was retarded in all the German children. It was known that these children were also short for their ages, and it was thought that it would be of interest to see how their bone age was related to their height age at the beginning and end of the investigation. The height age of each child was accordingly calculated, using the standards of O'Brien, Girshick and Hunt (1941) and expressed in the same 6-monthly

increments as the hand age. Table 3 has been constructed in the same way as Tables 1 and 2 and shows the means of the differences between the height ages and the chronological ages of the British children and also of the Duisburg and Vohwinkel children at the beginning and end of the year of experimental feeding.

TABLE 3

Means of the differences between the height ages and the chronological ages of the British children and of the Duisburg and Vohwinkel children at the beginning and end of the year of experimental diets

(*All bread groups together*)

ildren	Girls: No. in sample	Girls: Difference between height age and chronological age, Mean (with standard error) (months): Beginning	Girls: End	Girls: Increase in height age in excess of that expected during 12 months, Mean (with standard error) (months)	Boys: No. in sample	Boys: Difference between height age and chronological age, Mean (with standard error) (months): Beginning	Boys: End	Boys: Increase in height age in excess of that expected during 12 months, Mean (with standard error) (months)
ish	48	+ 5·1 ±2·6			54	+ 3·0 ±2·1		
iburg	50	−11·3 ±1·7	−5·6 ±2·0	+5·7 ±0·8	56	−12·9 ±1·6	−8·6 ±1·7	+4·3 ±0·6
winkel	27	− 9·3 ±2·3	−4·4 ±2·5	+4·9 ±0·9	30	−11·8 ±2·3	−7·4 ±2·6	+4·4 ±0·9

From this it may be seen that the height ages of the British girls and boys were slightly in advance of their chronological ages, whereas the height ages of the German children were initially 9–13 months behind their chronological ages. It will be recalled that over the year of experimental feeding the mean increase in bone age of the hands of the German children varied from 17 to 19 months. During this period the mean increase in height age of all the German children was about 17 months. Thus the mean gain in hand age was very similar to the gain in height age.

The correlation between bone age and height is brought out well in Fig. 5, which shows the regression lines correlating bone age of the hands with height. It may be seen that there was no significant difference between the lines for any of the series of boys, and that the lines for the German boys did not differ from those for the British control group. The lines obtained for the Duisburg and Vohwinkel girls at the beginning of the investigations did not differ significantly from the lines for the corresponding series at the end of the year, but they differed significantly from the line for the British girls. The shorter British girls had, on the average, a higher hand age than the shorter German girls, which is surprising as the British girls of comparable height tended to be younger.

When the heights at which the centres of ossification appeared in the British and German children were compared in the same way as the ages at which the centres became ossified, it was found that the height at which the various epiphysial centres appeared was much more similar from one series of children to another than was the age. In other words, the failure to grow in height was paralleled by a corresponding retardation in epiphysial development. This is in agreement with the findings of Stettner (1920–21), who observed a correlation between epiphysial development and height.

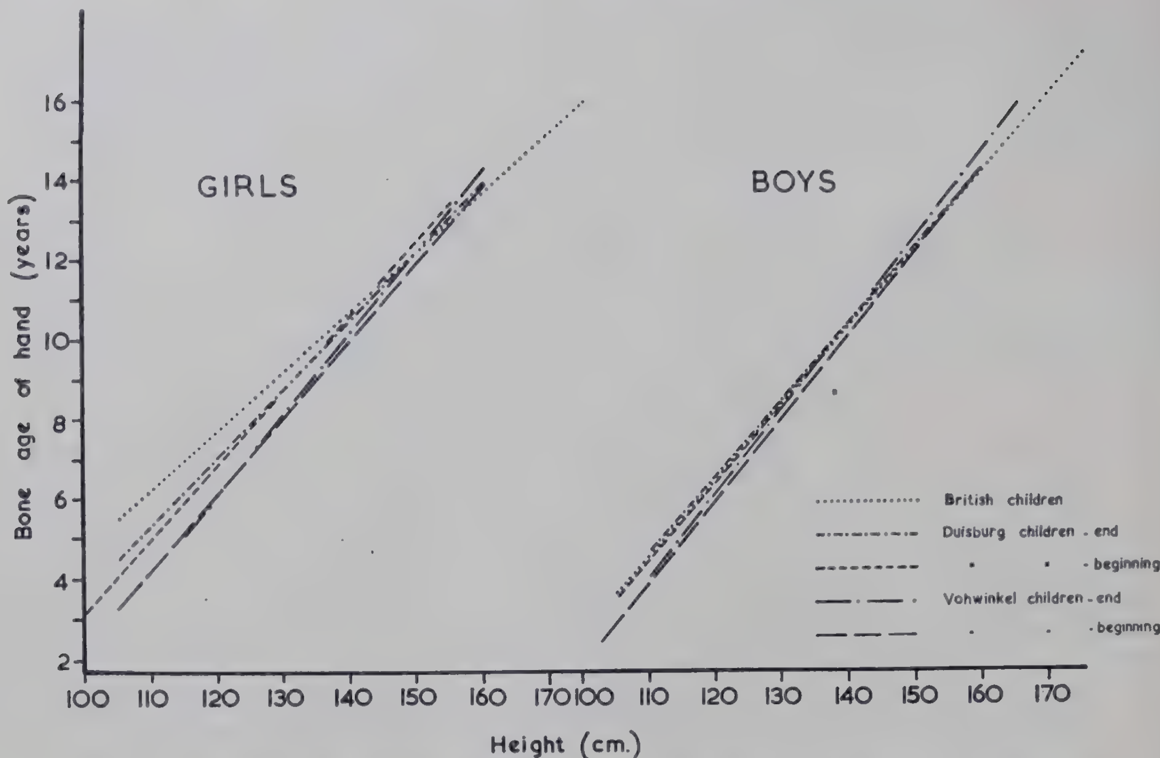

FIG. 5. The correlation between the bone age of the hand and height for all groups of children.

Discussion

The results of the radiological investigations described above have shown that, whatever standards are used for the assessment of ossification, the development of the bones of the children in the German orphanages was initially behind that of the bones of the British children examined in this country and of healthy children in America. It is probable that the retardation in their skeletal development was due to undernutrition, and this view accords with the findings of other workers who have X-rayed the bones of undernourished children (Grande Covian and Rof Carballo, 1944; Abbott, Townsend, French and Ahmann, 1950). It is widely recognized that epiphysial development is influenced by the amounts of vitamin D and calcium in the diet, but, since none of the German children showed clinical or radiological evidence of rickets or osteoporosis, it is probable that their diets had previously contained sufficient amounts of vitamin D and calcium for their slower rate of growth. MacNair and Roberts (1938) have shown that the ossification of the bones of children in an orphanage having "a mediocre institutional diet" developed less rapidly than that of others who were having the same diet supplemented with 16 oz. of milk a day (made up from evaporated milk). Irradiation of the milk was without greater effect, but this increased the vitamin D in the daily milk by only 70 units. MacNair (1939) made a further study in another orphanage, where the basal diet was much better than in the first, and included $1\frac{1}{2}$–2 pints of milk a day. She gave a dessertspoonful of cod liver oil to half the children and found that bone development was somewhat greater in these children than in the control group who received no supplement. The diets at the two orphanages in Germany contained very little milk, but the children in both homes were provided with 1–2 g. of calcium and 1,000 I.U. of vitamin D a day. There seems no doubt that these or other constituents of the diets were sufficient to enable bone development to keep pace with the rapid growth in height and weight.

Summary

1. The skeletal development of 50 girls and 56 boys, aged 5–14 years, at the orphanage at Duisburg and of 27 girls and 30 boys, aged 5–14 years, at the orphanage at Vohwinkel was assessed at the beginning and end of a year of experimental feeding and compared with that of a group of healthy British children of similar ages.

2. It was found that initially the ossification of the hands and knees and the epiphysial development of the German children was retarded as compared with the British children and with healthy children in the United States.

3. Over the year of experimental feeding:

(a) The ossification of the hands of the German children in both orphanages increased by significantly more than the expected increment over a period of 12 months.

(b) The ossification of the knees increased by just over a year.

(c) The epiphysial development of the bones of the German children tended to improve but did not reach that of the British children.

(d) The ossification of the hand and the epiphysial development of the German children increased in parallel with their growth in height.

References

Abbott, O. D., Townsend, R. O., French, R. B. and Ahmann, C. F. (1950). Carpal and epiphysial development. *Amer. J. Dis. Child.*, **79**, 69.

Behrendsen, (1897). Studien über die Ossifikation der menschlichen Hand vermittels des Röntgen'schen Verfahrens. *Dtsch. med. Wschr.*, **23**, 433.

Davies, D. A. and Parsons, F. G. (1927). The age order of the appearance and union of the normal epiphyses as seen by X-rays. *J. Anat.*, **62**, 58.

Engelbach, W. and McMahon, A. (1924). Osseous development in endocrine disorders. *Endocrinology*, **8**, 1.

Flecker, H. (1942). Time of appearance and fusion of ossification centres as observed by roentgenographic methods. *Amer. J. Roentgenol.*, **47**, 97.

Flory, C. D. (1936). Osseous development in the hand as an index of skeletal development. Monograph 3, Society for Research in Child Development, Washington D.C., National Research Council, 1936. Quoted by Todd, 1937.

Francis, C. C. (1940). Appearance of centres of ossification from 6 to 15 years. *Amer. J. phys. Anthrop.*, **27**, 127.

Francis, C. C. and Werle, P. P. (1939). Appearance of centres of ossification from birth to 5 years. *Amer. J. phys. Anthrop.*, **24**, 273.

Freeman, F. N. and Carter, T. M. (1924). A new measure of the development of the carpal bones and its relation to physical and mental development. *J. educ. Psychol.*, **15**, 257.

Gates, A. I. (1924). The nature and educational significance of physical status and of mental, physiological, social and emotional maturity. *J. educ. Psychol.*, **15**, 329.

Grande Covián, F. and Rof Carballo, J. (1944). Diet and development in children. 3. Radiographic study of skeletal development in a group of children of school age. *Rev. clin. espanola*, **12**, 234.

Greulich, W. W. and Pyle, S. I. (1950). *Radiographic atlas of skeletal development of the hand and wrist*. Stanford University Press, California.

Harris, H. A. (1933). *Bone growth in health and disease*. Oxford University Press; Mitford, London.

Heimann, A. and Potpeschnigg, (1907). Über die Ossifikation der kindlichen Hand. *Jb. Kinderheilk.*, **65**, 437.

Lurie, L. A., Levy, S. and Lurie, M. L. (1943). Determination of bone age in children. *J. Pediat.*, **23**, 131.

MacNair, V. (1939). Effect of a dietary supplement on ossification of the bones of the wrist in institutional children. Effect of a cod liver oil supplement. *Amer. J. Dis. Child.*, **58**, 295.

MacNair, V. and Roberts, L. J. (1938). Effect of a milk supplement on the physical status of institutional children. Ossification of the bones of the wrist. *Amer. J. Dis. Child.*, **56**, 494.

O'Brien, R., Girshick, M. A. and Hunt, E. P. (1941). Body measurements of American boys and girls for garment and pattern construction. *Misc. Publ. U.S. Dep. Agric.* No. 366, Washington, D.C.

Poland, J. (1898). *Traumatic separation of the epiphyses*. Smith, Elder and Co., London.

PRYOR, J. W. (1906). Ossification of the epiphyses of the hand. X-ray method. *Bulletin of the State University of Kentucky, Transylvania Company, Lexington, Kentucky.*

PRYOR, J. W. (1925). Time of ossification of the bones of the hand of the male and female and union of epiphyses with the diaphyses. *Amer. J. phys. Anthrop.*, **8**, 401.

ROTCH, T. M. (1909). A study of the development of the bones in childhood by the roentgen method, with the view of establishing a developmental index for the grading of and protection of early life. *Trans. Ass. Amer. Phys.*, **24**, 603.

SAWTELL, R. O. (1929). Ossification and growth of children from one to eight years of age. *Amer. J. Dis. Child.*, **37**, 61.

SHELTON, E. K. (1931). Roentgenographic studies in normal osseous development. *J. Amer. med. Ass.*, **96**, 759.

SIEGERT, F. (1935). *Atlas der normalen Ossifikation der menschlichen Hand.* Georg Thieme, Leipzig.

STETTNER, E. (1920–21). Über die Beziehungen der Ossifikation des Handskeletts zu Alter und Längenwachstum bei gesunden und kranken Kindern von der Geburt bis zur Pubertät. *Arch. Kinderheilk.*, **68**, 342.

STETTNER, E. (1921). Über die Beziehungen der Ossifikation des Handskeletts zu Alter und Längenwachstum bei gesunden und kranken Kindern von der Geburt bis zur Pubertät. *Arch. Kinderheilk.*, **69**, 27.

STETTNER, E. (1935). Normalia. Normaldaten für die Entwicklung der Knochenkerne. *Kinderärztliche Praxis*, **6**, 105.

TODD, T. W. (1932). Roentgenographic appraisement of developmental growth in the skeleton. White House Conference on Child Health and Protection. Growth and Development of the Child. Part IV, Appraisement of the Child, p. 258. The Century Company, New York.

TODD, T. W. (1937). *Atlas of Skeletal Maturation. The Hand.* Henry Kimpton, London.

VON RANKE, H. (1896). Discussion following "Über die Verwerthung der Röntgen'schen Strahlen in der Chirurgie" by Herr Angerer. *Münch. med. Wschr.*, **43**, 688.

VON RANKE, H. (1898). Die Ossifikation der Hand unter Röntgenbeleuchtung. *Münch. med. Wschr.*, **45**, 1365.

APPENDIX E: DENTAL CARIES AND TOOTH STRUCTURE

by Helen Mellanby

EARLY in January, 1948, the first detailed dental examinations were made of the children in the two orphanages. By this time the dietary studies at Duisburg had already been running for 6 months, but those at Wuppertal–Vohwinkel had not yet started. The final examinations took place in November and December, 1948, towards the close of the experimental period at Vohwinkel, and 4 months after the work at Duisburg was finished. The children at Duisburg, however, continued to eat the same type of diet, and it seemed of value to record the findings. Only those children who were examined on both occasions are included in the presentation of the results.

The teeth of 39 children belonging to another orphanage in Wuppertal were also inspected for dental caries, first in July, 1948, and again 5 months later, in

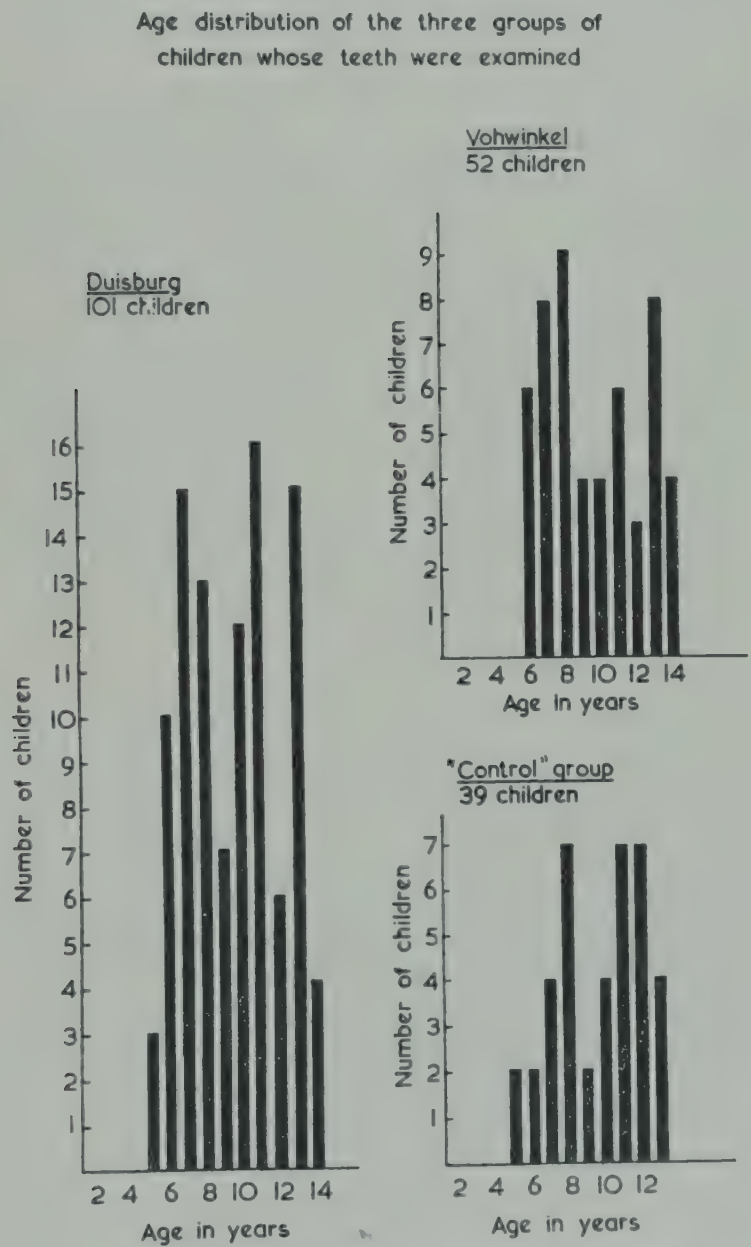

FIG. 1. Age distribution of children at the first examination.

November. These children were receiving only their German rations, without any supplements of calcium or vitamin D, and they may be regarded as 'controls'.

The age distribution of all three series of children at the first examination is shown in Fig. 1.

Methods

The standards adopted for tooth structure and dental caries were those used by May Mellanby and her colleagues in their surveys of London schoolchildren (Mellanby and Coumoulos, 1944, 1946; Mellanby, M. and Mellanby, H., 1948, 1951, 1952; Mellanby, H. and Mellanby, M., 1950). The kind of structural defect specified here has been termed 'M-hypoplasia' (King, 1940) to distinguish it from other types. It was assessed by the method devised by May Mellanby, (1934, 1937), which consists of rubbing a right-angled probe on the labial surface of each tooth, the teeth being graded according to the degree of roughness felt. The structure of the teeth was recorded at the first inspection only.

Caries was estimated with an illuminated mirror and rat-tailed dental explorer, its extent in each tooth being graded as C_1, C_2 or C_3 according to the size of the cavity. From a comparison of the first with the final charting the increase in the incidence of the disease, as well as the spread of that which had been originally present, was calculated. Some of the children had mixed deciduous and permanent dentitions, so that it would have been possible for some individuals to reduce their caries score by shedding carious deciduous teeth during the investigation. To allow for this contingency, all deciduous teeth shed after the first inspection were carried forward with their caries score (C_0, C_1, C_2 or C_3) to the final examination. The few teeth extracted during the intervals between the examinations at Vohwinkel were counted as severe caries (C_3) after extraction, and in most instances this had also been their initial caries figure. Permanent teeth were treated in the same way, but those erupting after the first examination were excluded from the totals.

Results

TOOTH STRUCTURE

The results of the examination of the deciduous and permanent teeth for surface structure are given in Tables 1 and 2. The deciduous teeth of the children both at Duisburg and Vohwinkel were of reasonably good structure, nearly half of them showing no M-hypoplasia. Since the children were aged 5–14 years the teeth included a larger proportion of deciduous molars (which are on the whole rougher than incisors) than the teeth of the 5-year old children forming the material of the London surveys. The figures for these German children also compare favourably with those of children in English orphanages (Coumoulos and Mellanby, 1947; Mellanby, M. and Mellanby, H., 1951).

The permanent teeth were of better structure at Vohwinkel than at Duisburg, some 13·6 per cent being free of hypoplasia at Vohwinkel as compared with 9·8 per cent at Duisburg; the corresponding figure for 14-year old L.C.C. pupils was 23·6 per cent in 1947 and 30·6 per cent in 1950 (Mellanby, M. and Mellanby, H., 1952). An unusually high proportion of the permanent teeth of the children

TABLE 1

Structure of deciduous and permanent teeth in 101 children at Duisburg

Grade of M-hypoplasia	Deciduous		Permanent	
	Number of teeth	Percentage	Number of teeth	Percentage
Hy_0 No hypoplasia	372	47·8	123	9·8
Hy_1 Slight hypoplasia ..	232	29·8	632	50·5
Hy_2 Definite hypoplasia ..	151	19·4	326	26·1
Hy_3 Severe hypoplasia ..	10	1·3	41	3·3
Gross or text-book hypoplasia	13	1·7	129	10·3
Totals	778		1,251	

TABLE 2

Structure of deciduous and permanent teeth in 52 children at Vohwinkel

Grade of M-hypoplasia	Deciduous		Permanent	
	Number of teeth	Percentage	Number of teeth	Percentage
Hy_0 No hypoplasia	210	45·7	98	13·6
Hy_1 Slight hypoplasia ..	148	32·2	396	55·1
Hy_2 Definite hypoplasia ..	79	17·2	205	28·5
Hy_3 Severe hypoplasia ..	11	2·4	4	0·6
Gross or text-book hypoplasia	12	2·5	16	2·2
Totals	460		719	

at Duisburg (10·3 per cent) showed gross hypoplasia. The corresponding figures for the London children quoted above were 1 per cent in 1947 and 2·8 per cent in 1950. The incidence of gross hypoplasia at Vohwinkel, however, was only 2·2 per cent of all permanent teeth, which corresponds more closely with the average figure for children in other parts of the world.

An additional feature of interest was mild dental fluorosis among 17 of the Duisburg children. This may be partly responsible for the small increase of caries at this home (see below). It is not known from what source the fluorine came as no information was available about the location and water supplies of the previous dwellings of the children. The water supply of the orphanage was analysed by Professor M. M. Murray, and was shown to contain only a trace of fluorine at the time when these investigations were made.

DENTAL CARIES

Table 3 shows the percentage of the total number of deciduous and permanent teeth which were recorded as carious when 101 children at Duisburg were

TABLE 3

Increase in the incidence and extent of dental caries at Duisburg

101 *children, age limits* 5 *years* 9 *months to* 14 *years* 11 *months in February*, 1948. *Average age* 10 *years* 5 *months. Re-examined* 10 *months later.*

Deciduous teeth shed during the interval have been included in the totals, but permanent teeth erupting after the first inspection have been omitted. There were no extractions.

Number of teeth	Percentage carious at 1st inspection	Percentage carious at 2nd inspection (10 months later)	Percentage increase in caries	A.C.F.*	
				1st inspection	2nd inspection
Deciduous 807 ..	18·8	20·2	1·4	0·25	0·28
Permanent 1,440 ..	12·9	14·8	1·9	0·19	0·22
All teeth 2,247 ..	15·0	16·7	1·7	0·21	0·24

* A.C.F. = Average Caries Figure = $\dfrac{\text{Number of teeth graded } C_1 + \text{twice number of teeth graded } C_2 + \text{three times number of teeth graded } C_3}{\text{Total number of teeth}}$

examined in January 1948, and again 10 months later. It is not possible to say what would have been the caries increment for these children had they continued with their ordinary orphanage diet, but it can be stated with certainty that the increase in both incidence and extent during the 10 months of observation was very small. Only 1·7 per cent of new teeth were attacked, which is equivalent to 1 tooth a year for every 3 children. The spread of existing caries was also small as judged by the rise in the average caries figure (A.C.F., see Table 3) from 0·21 to 0·24.

The incidence of caries at the beginning and end of the investigation at Vohwinkel is given in Table 4; these children had originally more caries in their

TABLE 4

Increase in the incidence and extent of dental caries at Vohwinkel

52 *children, age limits* 6 *years to* 14½ *years in February*, 1948. *Average age* 10 *years. Re-examined* 10 *months later.*

Deciduous teeth shed or extracted during the interval have been included in the total, but permanent teeth erupting after the first inspection have been omitted.

Number of teeth	Percentage carious at 1st inspection	Percentage carious at 2nd inspection (10 months later)	Percentage increase in caries	A.C.F.*	
				1st inspection	2nd inspection
Deciduous 464 ..	17·9	21·8	3·9	0·32	0·39
Permanent 721 ..	15·4	19·3	3·9	0·24	0·34
All teeth 1,185 ..	16·4	20·3	3·9	0·27	0·36

* A.C.F. = Average Caries Figure (see Table 3).

permanent teeth than the children at Duisburg, but the figure for the deciduous teeth was about the same at the two orphanages. The increase in carious teeth during the 10 months was higher at Vohwinkel than at Duisburg (3·9 per cent as compared with 1·7 per cent) but even so it cannot be regarded as large. It is the equivalent of about 1 tooth per child per year and was similar to that recorded for a group of Birmingham orphanage children observed for 3 years in 1928–31 (Committee for the Investigation of Dental Disease, 1936). Stones, Lawton, Bransby and Hartley (1949) give the average number of new carious teeth per child per year as 1·3 in a large Liverpool orphanage during the years 1942–4. The spread of existing caries, as measured by the A.C.F., showed an increase from 0·27 to 0·36, which was also greater than at Duisburg.

The teeth of the 39 children belonging to the 'control' orphanage group in Wuppertal had about the same total amount of caries at the beginning as those of the children at the other two orphanages (Table 5), but the percentage increase was 3·6 per cent of all teeth over a period of 5 months or approximately 1·8

TABLE 5

Increase in the incidence and extent of dental caries in a 'control' group of German orphanage children

39 *children, age limits* 5 *years to* 14 *years in July*, 1948. *Average age* 10 *years* 1 *month. Re-examined* 5 *months later.*

Deciduous teeth shed during the interval have been included in the totals but permanent teeth erupting after the first inspection have been omitted.

Number of teeth	Percentage carious at 1st inspection	Percentage carious at 2nd inspection (5 months later)	Percentage increase in caries	A.C.F.*	
				1st inspection	2nd inspection
Deciduous 272 ..	23·2	26·1	2·9	0·47	0·54
Permanent 542 ..	10·5	14·4	3·9	0·16	0·23
All teeth 814 ..	14·7	18·3	3·6	0·27	0·34

* A.C.F. = Average Caries Figure (see Table 3).

teeth per child per year. The period of observation was too short for any definite conclusions to be drawn, but the caries increase for both deciduous and permanent teeth appeared to be considerable for such a short time.

Discussion

Figures are not available for the expected annual increase of caries among German children living in their own homes, but figures for schoolchildren in other countries provide some data for comparison. Anderson, Williams, Halderson, Summerfeldt and Agnew (1934) reported the average yearly increase in caries for Toronto schoolchildren as 3 cavities per child, and Klein and Palmer (1940) found one new cavity in permanent teeth per child per year among schoolchildren in Haggerstown, U.S.A.

Children brought up in residential homes have been shown by a number of workers to have less caries than those of a similar economic level living at home (Anderson *et al.*, 1934; Collett, 1935; Schiotz, 1939; King, 1946; Boyd and Cheyne, 1947; Coumoulos and Mellanby, 1947; Knowles, 1948; Hadjimarkos and Storvick, 1949; Stones, Lawton, Bransby and Hartley, 1950; Mellanby, M. and Mellanby, H., 1951). This has been variously attributed to the influence of a regimented life, to a low sugar intake, or to the superior calcifying qualities of institutional diets compared with the possibly more varied, but in many cases less nutritionally adequate, food which a child receives when living with its family.

The teeth of orphanage children, on the other hand, tend to have slightly worse structure than those of children living in their own homes (Coumoulos and Mellanby, 1947; Mellanby, M. and Mellanby, H., 1951; Mellanby, H., unpublished results). The low incidence of caries among institutional children, therefore, suggests that the favourable influence must be post-eruptive.

There is no doubt that the increase in incidence of caries was small at the two German orphanages taking part in the investigations described in this report. The food supplements at these two homes, consisting as they did of large amounts of bread and jam, with additional refined sugar and margarine at Vohwinkel, might well have been expected by many workers to have increased considerably the prevalence and extent of dental caries, but there is no evidence that they did so. The results suggest, however, that the supplements of calcium and vitamin D which all the children in both orphanages received (pp. 13, 15) were sufficient to offset any possibly harmful effects on the teeth of other factors in the diets.

Summary

1. The deciduous and permanent teeth of 101 children at Duisburg and of 52 children at Vohwinkel who took part in the investigations described in this report were examined for structure (M-hypoplasia) and incidence of caries in January, 1948, and again in November, 1948.

2. The surface structure of deciduous teeth, as assessed by May Mellanby's probe method, was reasonably good in both homes. The permanent teeth at Duisburg were less well formed than those at Vohwinkel and contained an unusually large percentage of 'gross' hypoplastic specimens.

3. The increase in the incidence of caries was not large at either home, being equivalent to 1 tooth for every 3 children per year at Duisburg and 1 tooth per child per year at Vohwinkel.

4. The children in another orphanage in Wuppertal, who were receiving only their German rations, were also inspected twice, at an interval of 5 months. The percentage increase in caries was higher than it was in either home taking part in the dietary investigations.

References

Anderson, P. G., Williams, C. H. M., Halderson, H., Summerfeldt, C. and Agnew, R. G. (1934). The influence of vitamin D in the prevention of dental caries. *J. Amer. dent. Ass.*, **21**, 1349.

Boyd, J. D. and Cheyne, V. D. (1947). Epidemiologic studies in dental caries; The incidence of caries among institutionalised children. *J. Pediat.*, **31**, 306.

Collett, A. (1935). Tennene i et barnehjem og tennene i et daghjem i Oslo. *Tidsskr. Norske Laegeforem*, 22.

Committee for the Investigation of Dental Disease (1936). The influence of diet on caries in children's teeth. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 211.

COUMOULOS, H. and MELLANBY, M. (1947). Dental conditions of five-year old children in institutions and private schools compared with L.C.C. schools. *Brit. med. J.*, **1**, 751.
HADJIMARKOS, D. M. and STORVICK, C. A. (1949). Geographic variations of dental caries in Oregon. Dental caries among institutionalised children and the possible influence of certain ecological factors on its incidence. *J. dent. Res.*, **28**, 594.
KING, J. D. (1940). Dental disease in the island of Lewis. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 241.
KING, J. D. (1946). Dental caries. Effect of carbohydrate supplements on susceptibility of infants. *Lancet*, **1**, 646.
KLEIN, H. and PALMER, C. E. (1940). Studies on dental caries. The prevalence and incidence of dental caries experience, dental care, and carious defects requiring treatment in High School children. *U.S. Pub. Hlth Rep.*, **55**, 1258.
KNOWLES, E. M. (1948). A survey of the dental condition of children in day and residential wartime nurseries. *Brit. dent. J.*, **84**, 119.
MELLANBY, H. and MELLANBY, M. (1950). Dental structure and caries in 5-year old children attending London County Council schools. *Brit. med. J.*, **1**, 1341.
MELLANBY, M. (1934). Diet and the teeth. An experimental study. Part III. The effect of diet on dental structure and disease in man. *Spec. Rep. Ser. med. Res. Coun., Lond.*, No. 191.
MELLANBY, M. (1937). The role of nutrition as a factor in resistance to dental caries. *Brit. dent. J.*, **62**, 241.
MELLANBY, M. and COUMOULOS, H. (1944). The improved dentition of 5-year old London school children. Comparison between 1943 and 1929. *Brit. med. J.*, **1**, 837.
MELLANBY, M. and COUMOULOS, H. (1946). Teeth of 5-year old London school children (second study). Comparison between 1929, 1943 and 1945. *Brit. med. J.*, **2**, 565.
MELLANBY, M. and MELLANBY, H. (1948). The reduction in dental caries in 5-year old London school children (1929–1947). *Brit. med. J.*, **2**, 409.
MELLANBY, M. and MELLANBY, H. (1951). A further study of the teeth of 5-year old children in residential homes and day schools. *Brit. med. J.*, **1**, 51.
MELLANBY, M. and MELLANBY, H. (1952). Dental disease among 14-year old London school children in 1947 and 1950. *Brit. med. J.*, **1**, 7.
SCHIOTZ, E. H. (1939). Dental caries and nutrition (rural Norway orphanages). *Brit. dent. J.*, **66**, 57.
STONES, H. H., LAWTON, F. E., BRANSBY, E. R. and HARTLEY, H. O. (1949). The effect of topical applications of potassium fluoride and of the ingestion of tablets containing sodium fluoride on the incidence of dental caries. *Brit. dent. J.*, **86**, 263.
STONES, H. H., LAWTON, F. E., BRANSBY, E. R. and HARTLEY, H. O. (1950). Dental caries and length of institutional residence. *Brit. dent. J.*, **89**, 199.

Printed in Great Britain under the authority of Her Majesty's Stationery Office
by John Wright & Sons Ltd., at the Stonebridge Press, Bristol.

K16.

AO 5230. Wt. 2643. 1054. 10/54. J. W. & Sons Ltd. 518.